Carbon nanotubes: Impacts and behaviour in the terrestrial ecosystem - A review

Clarisse Liné [a, b], Camille Larue [a], Emmanuel Flahaut [b, *]

[a] ECOLAB, Université de Toulouse, CNRS, INPT, UPS, France
[b] CIRIMAT, UMR CNRS 5085/LCMI, Centre Inter-universitaire de Recherche et d'Ingénierie des Matériaux, Université Paul-Sabatier, F 31062, Toulouse Cedex 4, France

A B S T R A C T

For more than twenty years, nanotechnologies have arisen a huge interest and are used in numerous fields. Carbon nanotubes (CNTs) are one of the most used nanomaterials thanks to their excellent optical, mechanical, electrical and thermal properties. All along their lifecycle, CNTs may be spread in the environment during production, use, destruction, reuse or potential accidents in production units or during transportation. For this reason, it is essential to evaluate their behaviour and potential impacts on ecosystems and particularly on the terrestrial ecosystem. After a brief summary of CNT properties, synthesis methods, and applications as well as detection and characterisation techniques, this review will focus on impacts of CNTs on the terrestrial ecosystem, discussing their behaviour in soil, plants and interactions with other pollutants as well as their impacts on soil microbiota, macrobiota and plants.

Keywords:
Carbon nanotubes
Impacts
Behaviour
Terrestrial ecosystems
Soil
Plants
Soil microorganisms
Soil macroorganisms

Contents

Abbreviations: CBNMs, Carbon Based Nanomaterials; UV–vis–NIR, Ultraviolet–Visible and Near-Infrared Spectroscopy; TEM, Transmission Electron Microscopy; AFM, Atomic Force Microscopy; SEM, Scanning Electron Microscopy; GC-MS, Gas Chromatography-Mass Spectrometry; GC-ECD, Gas Chromatography with Electron Capture Detector; EDS, Energy-Dispersive X-ray Spectroscopy; TGA, Thermo-Gravimetric Analysis; BET, Brunauer-Emmett-Teller; qPCR, Real-Time Polymerase Chain Reaction; Integrated PC/PT scanning cytometry, Integrated PhotoThermal and PhotoAcoustic scanning cytometry; ICP-MS, Inductively Coupled Plasma Mass Spectrometry; FTIR, Fourier Transform Infrared Spectroscopy; N.I, Non Informed Information; QD, Quantum Dot; DDx, DDT (dichlorodiphenyltrichloroethane) + DDE (dichlorodiphenyldichloroethylene) + DDD (dichlorodiphenyldichloroethane); MS medium, Murashige and Skoog medium; SWCNH, Single Wall Carbon NanoHorns; ROS, Reactive Oxygen Species; SPAOM, small polar aromatic organic molecules.

* Corresponding author.
E-mail address: flahaut@chimie.ups-tlse.fr (E. Flahaut).

1. Introduction

For more than a decade, nanotechnologies are more and more investigated by industrials and scientists and used worldwide for applications thanks to their remarkable properties. The European Commission defined in 2011 a nanomaterial as "A natural, incidental or manufactured material containing particles, in an unbound state or as an aggregate or as an agglomerate and where, for 50% or more of the particles in the number size distribution, one or more external dimensions are in the size range 1 nm−100 nm" [1]. However, nanomaterial definition is different according to countries and to the field in which they are used. All definitions agree about the nanoscale dimensions but definitions differ on size distribution for example. This lack of global consensus is a serious challenge because it leads to legal uncertainty and differing regulatory for the same nanomaterial. The nanotechnology consumer products inventory (CPI) listed officially in 2014 more than 1800 consumer products containing nanoparticles worldwide. In less than ten years, the number of products containing nanoparticles increased by more than 3000% (54 products in 2005) [2].

Carbon-based nanomaterials are among the most used [2]. There are different types of carbon nano-objects such as fullerenes (3 dimensions < 100 nm), carbon nanotubes (2 dimensions < 100 nm, CNTs) and graphene and related materials (1 dimension < 100 nm). Since their discovery in 1991 by Iijima, they arose an extraordinary enthusiasm [3,4]. CNTs can be described as graphene sheets rolled over themselves to form (concentric) cylinders with a nanometric diameter. We can define three kinds of CNTs: single wall CNTs (SWCNTs), double wall CNTs (DWCNTs) with two concentric tubes and multi wall CNTs (MWCNTs) with more than two concentric tubes. CNT diameter varies from a few nanometers for SWCNTs to several tens of nanometers for MWCNTs. Their length is usually of a few micrometers. CNTs have remarkable optical, electrical, thermal, mechanical and chemical properties. They are used in numerous fields such as plastic additives, in batteries or some sporting goods [5].

It is essential to regulate production and uses of nanomaterials for a safe and sustainable future. So far there is no international agreement to supervise the production, use and commercialisation of nanomaterials. However, few countries started to monitor nanomaterials commercialised in their territories by using registers. In Europe, there is the European regulation for the recording, evaluation, authorization and restrictions about chemical substances (REACh). The recording and the authorization are compulsory for produced or imported nanomaterials with a volume of more than 100 tons. A new authorization protocol will be apply in 2018 for volumes between 1 and 100 tons, without toxicological data required. In theory, nanomaterials are covered by this regulation but practically they are often brought to the market without preliminary recording or monitoring. The first reason is that producers and distributors produce or import very rarely more than one ton per year, the threshold below which it is not compulsory to make a REACh recording. The second reason is that even if there is more than one ton per year, REACh does not oblige to record nanomaterials as new substances. Consequently, the recording gets an extension and the terms and conditions are simplified excluding for example ecotoxicological data. In France, a precursor in this domain, since January 1st, 2013, industrials and researchers have to declare annually the quantity, the properties and the uses of nanomaterials they produce or import in the R-Nano database handled by the ANSES (French Agency for Food, Environmental and Occupational Health & Safety) (L. 523-1 and L 523-3 of "Code de l'environnement" [6]). In Norway, since 2013, the national public agency of climate and pollution asks for identification of nanomaterials in the chemical product register. In Denmark, producers and importers have to record nanomaterials and products containing or releasing nanomaterials since 2014. Finally, in Belgium, since 2016 there is a royal decree concerning the placing on the market of manufactured nanomaterials.

In the USA, regulations for nanomaterials have been established by numerous organizations including the Environmental Protection Agency (EPA), Food and Drug Administration (FDA), and Consumer Product Safety Commission (CPSC). EPA is controlling nanomaterials by existing regulations of the Toxic Substances Control Act (TSCA) and Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) pursuant Significant New Use Rules (SNUR) of premanufacture notices (PMNs) of 13 chemicals, including CNTs and fullerenes. For nanomaterial manufacture and production, the manufacturers must inform the EPA with information about the nanomaterials within 90 days. For the FIFRA regulation, pesticide products containing nanomaterials must be registered. In Asia, the Japanese Council for Science, Technology and Innovation (CSTI) is paying attention to the new rules implemented in EU and USA. However, there is no legal control related to nanomaterial safety and environment so far. Anyway, Japanese Government is working with ministry of economy, trade and industry (METI) in order to collect information about the industry working with nanomaterials and to evaluate harmful effects of nanomaterials with the ministry of environment. Broadly speaking, scientists, associations and sanitary agencies are worried about the risks associated with nanomaterials and nanotechnology. However, industrials do not want regulatory framework because in the European and international market, nanotechnology is bringing jobs. So far, there is no strict regulation on nanotechnology. However, it is an international problem for environment, safety and health, it is thus essential to roll out international rules for their control.

All along their lifecycle, CNTs may be spread in the environment during production, use, destruction, reuse or potential accidents in production units or during transportation [7]. During their release, they can be subjected to physico-chemical modifications which may later modulate their potential toxic effects [8]. Toxicological studies evidenced that, CNTs present a potential risk for humans upon pulmonary exposure. CNT effects raise concerns because they can be compared to asbestos due to their fibre shape [9]. Asbestos caused a worldwide pandemia of disease in the 20th century such as asbestosis, mesothelomia, bronchogenic carcinoma, etc. [9]. For instance, Kasai et al. [10] studied the toxicity of MWCNTs with whole-body inhalation exposure in rats; they found that MWCNTs increased lung weight and inflammatory parameters of the exposed rats.

It is also essential to assess their behaviour and potential impacts on ecosystems. To date, the focus has been mainly on aquatic ecosystems rather than on the terrestrial ecosystems [11]. This review aims at summarizing the knowledge about behaviour and impacts of CNTs on the terrestrial compartment with a focus on plants. Our survey covered 71 studies on terrestrial ecosystems. The majority of the studies have been realized on plants (65%). Soil microorganisms and macroorganisms have been studied with respectively 14% and 17% of the studies. The less studied domain is the behaviour of CNTs in soil (in laboratory soil column) with only 4% of the mentioned articles. For plants, 46 studies have been published, with different culture conditions (Fig. 1a): most of the studies were based on plants exposed in a simplified media: hydroponics conditions (35%), filter paper (13%) and jellified medium (17%). Studies using soil exposure, representing the most relevant exposure scenario to mimic real environmental conditions, represent only 17% of the articles (15% in soil, 2% in sediment). The last part of the studies used *in vitro* tests on plant cells (16%). The exposure time is another parameter to take into account: among the 46 plant studies, 19% focus only on seeds (Fig. 1b). Most of the

studies were realized on seedlings (47%). Long-term exposure with adult plant represents 16% of the studies. Exposure during the entire life cycle, which represents the most realistic scenario, are only 2% of the cases. In total, 84 different plants were studied. 59% were dicotyledons and the rest monocotyledons. Different CNTs have also been studied: SWCNTs, MWCNTs, functionalized or not. MWCNTs are the most used for ecotoxicological studies on plants (more than 84%).

Scientists and industrials are getting more and more conscious of nanomaterial effects, at the same time they know the high potential of nanomaterials. Consequently, they are trying to find a compromise between these two aspects for example with the "safe by design approach". In this approach, physico-chemical parameters of nanomaterials are studied. Then, they are trying to find a way to reduce at the maximum the nanomaterial toxicity by playing with the different physico-chemical parameters [12].

In this review, general information on CNTs will be briefly reminded including CNT properties, their synthesis and their different applications. Then the issue of the detection and characterisation will be discussed. The other parts concern the environmental implications of CNTs with their release and potential exposure pathways, their fate and impacts on the soil system and finally the last part will focus on their fate and impacts on plants.

1.1. Carbon nanotube synthesis, properties and applications

High temperature preparation techniques were first used to produce CNTs such as arc discharge or laser ablation. Nowadays, these methods have been replaced by low temperature chemical vapour deposition (CVD) techniques [17]. With CVD techniques, the orientation, alignment, length, diameter, purity and density of CNTs can be controlled precisely. Other less common techniques can also be used for CNT synthesis such as liquid pyrolysis and bottom-up organic approaches [18]. Whatever CNT preparation method used, they always contain impurities, most of them corresponding to residual catalyst, but other unwanted carbon species are usually also present to some extent such as disorganised carbon. These impurities have to be chemically treated in order to be eliminated. They can be washed using concentrated acids such as hydrochloric acid or nitric acid. As-produced CNTs are hydrophobic, and thus obtaining a homogenous suspension of CNTs is challenging. To increase their hydrophilicity, CNTs can be functionalized by modification of the external wall. There are mainly two types of functionalisations. The first one, and the most used, is the covalent functionalisation using oxidising treatments which damage the outer wall of the CNTs while grafting oxygen-containing chemical groups. Covalent functionalisation implies strong treatments such as heating with acids, which are damaging CNTs. Consequently, functionalized CNTs are shorter than untreated ones. The second functionalisation is non covalent, and based on the adsorption of a surfactant to obtain a more homogeneous suspension of CNTs. Numerous dispersants/surfactants have been used in the literature. In order to work with living organisms it is required to use non-toxic dispersants. A sap exudate called Arabic gum can be used to disperse CNTs in suspension [19,20]. 0.25% (w/v) of Arabic gum is able to stabilize a suspension of 1 g/L of CNTs during one month at pH 5.5 [20]. Humic acid, one of the most important fraction of humus, can also be employed. 0.25% (w/v) of humic acid is also able to stabilize a suspension of 1 g/L of CNTs during one month at pH 7.6 [20]. Other dispersants can be used to disperse CNTs such as gallic acid, an aromatic organic compound common in plants [21], carboxymethylcellulose or tween 20, a non-ionic surfactant [19].

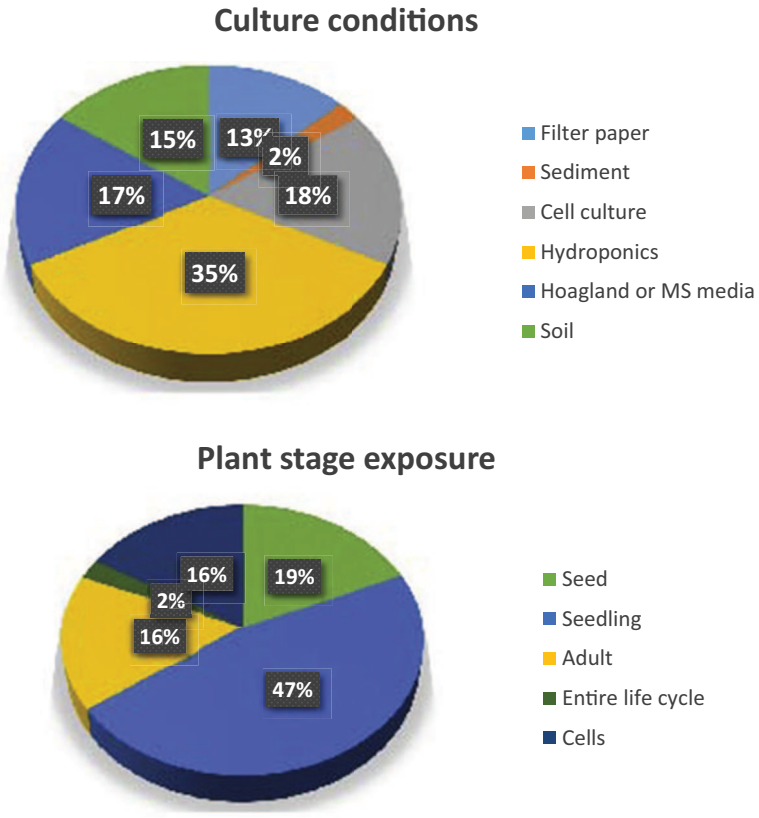

Fig. 1. Literature review of CNT impacts and behaviour on plants (culture conditions and plant stage exposure). Seedling represents plant after germination but still growing. Adult plants are plant which they reach adult height. (A colour version of this figure can be viewed online.)

Bile salts have also been used such as sodium cholate [22] or deoxycholic acid [23].

Due to their unique structure, CNTs display remarkable physical properties. From a mechanical point of view, they have an extraordinary flexibility despite their high rigidity [5]. The bending is reversible until a critical angle of 110° for a SWCNT [13]. CNTs are 100 times more resistant and 6 times lighter than steel [14]. They also have useful electrical properties: depending on their structure, they can behave like semiconductors or like metals. Thanks to their electronic properties and their good stability at high temperatures (up to 250 °C in air and >2600 °C in vacuum), CNTs can withstand extreme current densities (one order of magnitude more than copper) [15]. Regarding thermal properties, the low proportion of structural defaults and the cylindrical geometry of CNTs lead to high thermal conductivity along the axis of the nanotube, comparable to that of the isolated graphene sheet or high purity diamond ($\lambda \approx 6600$ W/m K) [16].

CNT uses and applications are increasingly numerous and varied: for example, they can be used as field emission sources for visible light [24] or X-rays [25]. Their high rigidity, their nanometric diameter and their flexibility allowed Dai et al. [26] to realize a tip for scanning probe microscope by gluing a CNT on the tip of an atomic force microscope [27]. Cheung et al. [28] have grown CNTs directly on the tip of an atomic force microscope allowing high resolution images [29]. CNTs can also be used in the composition of flat screen TV which allows a lower electricity consumption, a more intense luminosity and a larger range of operating temperatures [5]. They are used in batteries of electronic mobile devices allowing a better energy storage [30]. Most of the applications are however related to nanocomposite materials and paints in which CNTs usually bring some electrical conductivity in addition to an increase in the mechanical properties. CNTs are found in sport equipments such as tennis rackets, bicycle frames or golf clubs in order to make them lighter. They are also found in clothes making them more resistant and waterproof [2]. Alternatively, CNTs appear as a new alternative for biomedical applications, they can be efficient to transport and translocate therapeutic molecules [31], or even to fight cancer [32,33]. CNTs may also be used in agriculture. In plants, Serag et al. [34] investigated the ability of CNTs to penetrate the plant cell walls and most of the subcellular membranes to deliver payloads to specific cellular organelles in plants with the aim of increasing pesticide efficiency and thus their input in the environment. Wood could be reinforced with CNT fibres in order to increase its strength [36]. Due to numerous studies that report growth increase of plants after CNT application, CNTs are imagined to be used as fertilizers [35]. Finally, thanks to their high adsorption capacities, CNTs seem to be able to remove a diverse range of biological contaminants such as bacteria or viruses from water systems. They can also be used for the removal of chemical contaminants such as heavy metals or organics [36]. CNTs have been used as a sponge for oil during oil spill; they have great sorption capacity and the absorbed oil can be recovered by squeezing or be converted to heat by burning the oil within sponges [37].

1.2. Releases and potential exposure pathways

CNT spreading into the environment can occur following different routes. The release will usually be unintentional, with possible chronic and/or acute contaminations. Chronic dissemination corresponds to the contamination by low doses of CNTs, but over a long period of time. Direct release (chronic dissemination) has been considered as very low for most of the scenarios, except for tires [38]. For example, CNTs can be accumulated in soil due to the rubbing of CNT-containing tires on roads [38]. CNT have been found in the lungs of Parisian kids and this may be due to a production of such nanomaterials by car catalytic converters [39]. For most of the other life cycle stages (production, uses or end of life), releases can be possible but it is difficult to assess the real risk due to the lack of knowledge during and after waste management and recycling operations of nanomaterials [40].

Acute contamination corresponds to a high release but during a short period, for example during an accident in a production unit or during transportation [41]. Upadhyayula et al. [42] studied the life cycle assessment of products containing CNTs. They evidenced that the manufacturing stage of CNT containing products dominates the environmental impacts. Likewise, Nowack et al. [38] studied the potential release scenarios for CNTs during nanocomposite production. The authors concluded that release during manufacturing may be possible, but this is also the place where exposure can be best controlled.

It is important to mention here that if CNTs released from a material may be similar to the initial incorporated nanomaterial, either individual or agglomerated, CNTs functionalized by residual coating with the matrix material may also be observed [43]. The interactions between CNTs and their environment are also driven by the interface with the outer wall. The presence of residues of polymers, for example, may modify their wettability/hydrophobicity and thus influences directly their fate in water, soils and organisms.

So far, CNT concentration in the environment (as well as other nanomaterials) cannot be measured directly and the research in this domain can only rely on modelling results. Sun et al. [44] modelled the environmental concentrations of engineered nanomaterials including CNTs. In surface water, CNT concentration in 2014 was estimated to be around 0.36 ng/L, 6.74 μg/kg in sediment, 35 ng/kg in natural and urban soil, 11.7 μg/kg in sewage sludge treated soil and 0.02 ng/m^3 in the atmosphere. Gottschalk et al. [41] modelled flows and concentrations of 9 engineered nanomaterials in the Danish environment. Authors calculated that the primary sources of CNTs would be waste incineration plants (<1% of total primary sources), sewage treatment plant effluents and overflow (<1%), sewage treatment plant sludge (<1%) and production, manufacturing and consumption including untreated wastewater (99%). The primary recipients of CNTs were soils (91.2%), marine water (3.5%), freshwater (2.8%) and air (2.5%). According to their models, CNT concentrations in surface water of the Danish environment would be between 0.2 and 15 pg/L, in sediments (freshwater) between 0.1 and 5.6 μg/kg, between 18 and 75 ng/kg in agricultural soils, between 41 and 220 ng/kg in natural soils, between 71 and 290 ng/kg in urban soils and finally between 0.022 and 0.091 ng/m^3 in air.

The release can also be intentional, when for example CNTs are used for depollution (nanoremediation). Indeed, they have the potential to remove bacterial pathogens, natural organic matter and cyanobacterial toxins from water systems [36]. CNTs may also be used in plant protection or fertilizer products [35]. Numerous studies highlighted positive impacts of CNTs on plants, especially at rather low doses (see 5. Fate and impacts of carbon nanotubes on plants).

1.3. Detection and characterisation of carbon nanotubes in environmental matrixes

The detection and quantitative analysis of CNTs in biological samples is very complex because it is difficult to detect a specific form of carbon in a carbon based matrix. Sample preparation is often challenging in complex environments [48].

Many methods exist to detect CNTs, but apart from the use of isotopic labelling [45], it is generally difficult to analyse them both

qualitatively and quantitatively. However, this technic presents several constraints. It is expensive to synthetize CNTs with isotopic labelling like carbon 14 and authorization and adapted installations and equipment to work with carbon 14 are required. Labelling with carbon 13 is another alternative but it is not widespread. Microscopy techniques can be used such as scanning electronic microscopy (SEM) and transmission electronic microscopy (TEM) to determine the length, diameter and number of walls. TEM and SEM are also extensively used to localize CNTs in biological samples looking for fibre shaped structures (Fig. 2), however this technique does not provide a formal proof that the fibre is indeed a CNT. The specific surface area of a particle (m^2/g) is among the most important parameters to measure. It is even more crucial in ecotoxicology since Mottier et al. [46] evidenced that the surface area of carbon based nanomaterials is a dose metric more realistic than the size or the number of particles. There are different methods to measure the specific surface area of a particle but the most common is the Brunauer–Emmett–Teller (BET) method. This method is based on the Langmuir theory of physical adsorption of a gas monolayer on a solid [47]. However it can be used only in a nanomaterial powder (elimination of the bio-matrix). To analyse the chemical purity or rhe corona form around CNT, inductively coupled plasma (ICP) techniques are mainly used after a proper acidic digestion.

Herrero-Latorre et al. [49] also reviewed the different analytical methods for detection and characterisation of CNTs in environmental and biological samples. Raman spectroscopy can be used to give both qualitative and semi-quantitative information. Two bands in particular, the D one corresponding to sp^3-like carbon and the G one corresponding to sp^2 carbon, are mainly used. The band intensity (especially for the G band) can give information about the concentration and the orientation (polarization effects) of CNTs. The band surface gives indications about the quantity and can thus be used to estimate concentrations. The ratio (intensity or area) between the D and G bands allows measuring the proportion of defectuous carbon present in the sample [50]. There are several other techniques to characterize and detect CNTs such as atomic force microscopy (AFM) [51], dynamic diffusion of light, although this may not be well-suited for elongated and flexible nanomaterials such as CNTs [52], infrared spectroscopy [53] or photoluminescence [54]. Lutsyk *et* al. [55] recently proposed a new method using selective photoluminescent probes based on ionic complexes with organic dyes. Thermogravimetric analysis (TGA) may also be used; this is a thermal analysis technique measuring the mass variation of a sample *vs.* the applied temperature, in a controlled atmosphere. This technique is especially relevant in the context of the quantitative assessment of CNTs in complex environmental samples when coupled to other instruments such as mass-spectrometry as well as thermal optical transmittance/reflectance in order to differentiate organic and elemental carbon [56]. Microwave measurements have also been shown to be very sensitive for the specific quantification of CNTs in biological samples [57,58]. Conventional mass spectrometers have troubles to detect CNTs due to their large molecular weight. Chen *et* al. [59] overcome this problem by using the intrinsic carbon cluster fingerprint signal of the nanomaterials.

Smith et al. [60] used dark-field and hyperspectral imaging (HSI) to obtain spectral image of CNTs in monocytes. These technics are used for medicine purposes so far but it is possible to use them in ecotoxicology for the detection in environmental samples. Photothermal/photoacoustic imaging can also help to localize CNTs in plant leaves. Khodakovskaya et al. [61] used this method to analyse interactions between plants and CNTs.

Herrero-Latorre et al. [49] concluded that the characterisation of CNTs requires a wide range of analytical techniques because all the information usually cannot be obtained using one technique alone. Moreover there is a lack of standardized characterisation protocols which makes difficult the comparison of CNTs between studies. Nowadays the most common techniques are TEM, SEM, and Raman. The determination of CNTs in biological and environmental samples still constitutes one of the main challenges in the field.

1.4. Fate and impacts of carbon nanotubes on soil and related organisms

Depending on their length, diameter, functionalisation and on environmental conditions, CNTs may have a different behaviour in natural conditions [62].

Behaviour of CNTs in soils was little studied in the literature. However, this is essential to evaluate their potential impacts on terrestrial organisms. Jaisi and Elimelech [63] investigated the behaviour of carboxyl-functionalized SWCNTs in a column packed

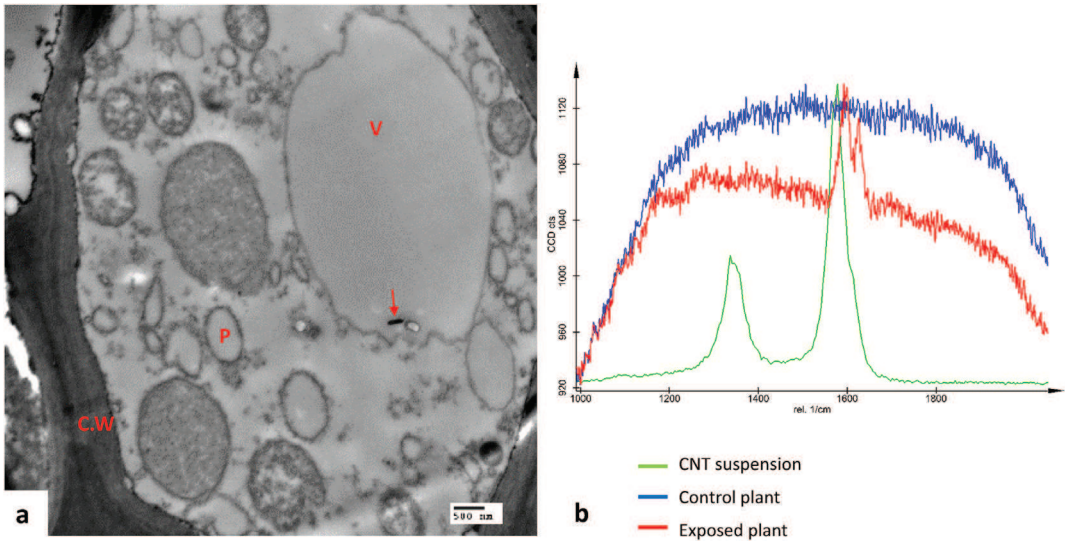

Fig. 2. a. TEM image of MWCNTs in wheat's roots; roots of wheat *(Triticum aestivum)* exposed to 100 mg/L of MWCNTs dispersed in gallic acid for 7 days; CNT is indicated by arrow; (C.W) cell wall; (P) plaste; (V) vacuole. b. Raman spectra of the CNT suspension, the control plant and the same exposed plant as the Tem image. (A colour version of this figure can be viewed online.)

with natural agricultural soil (fine sandy loam soil). They demonstrated that the deposition rate of SWCNTs was relatively high over a wide range of monovalent and divalent cation concentrations added to the soil solution (0.03–100 mM). Authors concluded that SWCNTs would not exhibit substantial transport and infiltration in soils because of effective retention by the soil matrix. Kasel et al. [64] studied the behaviour of ^{14}C-labeled MWCNTs in two different types of natural soils. There was a stronger sorption of CNTs on the silty loam soil compared to the loamy sand but the overall conclusion was that MWCNTs remained in the soil: more than 85% of the applied radioactivity was recovered in the soil fraction. Lu et al. [65] studied the behaviour of MWCNTs in 3 types of soils: positively charged MWCNTs were entirely retained in soils, while negatively charged CNTs broke through the soil column and were found in the outlet. They also demonstrated that soil texture, rather than organic matter, controlled MWCNT mobility. Cornelis et al. [66] reviewed the fate and bioavailability of engineered nanomaterials in soils. They concluded that some general trends can be deducted. Engineered nanomaterial bioavailability is higher in saturated, coarsely textured soil with high content of organic matter than in other soils. In unsaturated, finely textured soils with low organic matter content, nanomaterial bioavailability is expected to be low. CNT behaviour in soil media is dominated by the shape, structure and agglomeration state of CNTs in aqueous soil suspension, but also by the heterogeneity, particle size, porosity, nature and permeability of the soil. The agglomeration of CNTs with soil components and other micro and macroorganisms determine their impacts. In comparison with the aquatic compartment, CNTs in soil are more prone to hetero-agglomeration phenomena [67]. In water, CNTs are more likely to form homo-agglomerates but in sediments they seem to act like in soil [68]. Overall, most of CNTs seem to be retained in the soil fraction except in particular cases when negatively charged MWCNTs have been seen to leak out from the soil matrix.

CNTs may also interact with other pollutants present in the environment. Their large specific surface area can favour the adsorption of other pollutants (ionic species, organic molecules) and thus may influence the behaviour and the toxicity of CNTs and/or of co-pollutants [62]. Numerous authors studied the interaction between CNTs and other contaminants in aqueous solution [62], but here, we will focus on CNT interactions in soil. Shrestha et al. [69] studied the influence of MWCNTs on polycyclic aromatic hydrocarbons (PAH) bioavailability and toxicity to soil microbial community in alfalfa rhizosphere. They concluded that MWCNT influence on PAH varied according to the different soil types: in a soil with high organic matter content, MWCNTs increased the pyrene degradation [69]. MWCNTs generally minimized toxicity of highly bioavailable PAHs on microbial community. De la Torre-Roche et al. [70] studied the impacts of MWCNTs and C_{60} fullerenes on pesticide accumulation in agricultural plants. MWCNTs decreased chlordane and DDx (DDT + metabolites) accumulation across the 4 studied plants while C_{60} fullerenes completely suppressed DDx uptake but increased chlordane accumulation. There is a lack of information and understanding about CNT behaviour in soil and with other pollutants, more studies are needed.

CNT effects on soil microbial activity is controversial (Table 1) and was only little studied (12 articles in 10 years). However, the majority of these studies seem to conclude that CNTs decreased soil microbial activity [71–74]. Enzymatic activities of soil bacteria were repressed by both MWCNTs and SWCNTs: MWCNTs decreased enzymatic activities of two natural soils (sandy loam and loamy sand soils) at 500 mg/kg [71]. Likewise Jin et al. [72] found that SWCNTs lowered significantly enzyme activities of a natural sandy loam soil at concentrations between 30 and 300 mg/kg. In another study, bacterial soil community was affected by the presence of SWCNTs with a major impact after 3 days but bacteria recovered completely after 14 days [75]. Interestingly, Shan et al. [76] found that MWCNTs at low concentration (0.2 mg/kg) stimulated mineralization of an agricultural soil by bacteria. Ge et al. [74] made an interesting work about effects of MWCNTs compared to natural or industrial carbonaceous materials on soil microbial communities using long-term studies in dry soil. They found that MWCNTs reduced soil DNA diversity and altered bacterial communities after one year of exposure. These effects are similar to those observed for natural and industrial carbonaceous materials. There are not enough studies available so far to conclude about a possibly different impact between functionalized and unfunctionalized CNTs on soil microbial activities.

To date few studies (only 10) are available on the effects of CNTs on soil macroorganisms (Table 2). All of them focused on earthworms exposed in soil (natural or artificial) [77–79]. Some studies reported effects on whole organism endpoints such as reproduction or mortality [80,81] and two focused on sub-organism endpoints [81,82]. All studies agreed that CNT uptake by earthworms was rather low. CNTs can enter in earthworms by ingestion and phagocytosis through tissues but earthworms can also eliminate accumulated CNTs [79,83,84]. Consequently, their toxicity of CNTs was limited. No mortality was found in soil contaminated with MWCNTs even at high concentration (1000 mg/kg) but DNA damages and other sub-organism endpoint alterations were evidenced in earthworms at lower concentration (50 mg/kg). Finally, earthworms reproduction was affected by DWCNTs at concentrations between 50 and 500 mg/kg [85].

As soil is expected to be the main sink for CNTs, ecotoxicological risks of CNTs in terrestrial environment is of great concern. More studies focussing on CNT behaviour and impacts soil micro and macroorganisms are thus urgently needed.

1.5. Fate and impacts of carbon nanotubes on plants

CNTs can penetrate into the seeds of cabbage (*Brassica oleacera*) [86], rice (*Oryza sativa*) [87], tomato (*Solanum lycopersicum cv Micro-Tom*) [88,89], barley (*Hordeum vulgare* hybrid Robust), soybean (*Glycine* max hybrid S42-T4) [90] and maize (*Zea mays* hybrid N79Z 300 GT) in hydroponic conditions [91] (Table 3). Functionalized CNTs penetrated directly into the cells, not entering by phagocytosis mechanism [88–90,92]. When contamination occurred through root exposure, both functionalized and non-functionalized CNTs have been reported to penetrate (Fig. 3) [85,93,94]. Then, CNTs are translocated to the upper part of plants by sharing the vascular system with water and nutrients and they can be transported via transpiration (Fig. 3). CNTs are most of the time detected in stems, shoots, leaves and fruits of the plants although in low concentration [20,70,87,95–98]. Larue et al. [20] established that less than 0.05‰ of the applied MWCNT dose was translocated to the leaves of wheat (*Triticum aestivum*) and rapeseed (*Brassica napus*) using ^{14}C labeled MWCNTs. CNT seem to penetrate plant roots by osmotic pressure, capillarity forces, cell pores or symplastically (Fig. 3) [85,93,94]. Lin et al. [93] studied the intergenerational transfer of carbon nanomaterials (carbon nanoparticles C_{70} and MWCNTs 40–70 nm diameter) in rice (*Oryza sativa* L. ssp. japonica, cv Taipei 309). They concluded that carbon nanomaterials can pass to the progeny through seeds. Khodakovskaya et al. [61] found that CNTs could penetrate in chloroplasts through the lipid bilayer by lipid exchange. Serag et al. [99] proposed that MWCNTs can be taken up in plant protoplasts by endosome-escaping (Fig. 3). Moreover, short MWCNTs (<100 nm) were targeted to specific cellular sub-structures such as nucleus, plastids and vacuoles. Serag et al. [100] also reviewed that CNTs can penetrate plant cell walls, target specific organelles, probe protein-carrier activity and induce

Table 1
Studies on CNT behaviour and impacts on soil microorganisms.

Article	Soils/bacteria	Culture conditions	CNT used[a]	Concentrations	CNT characterisation	Effects
[71]	Sandy loam soil from a landscaped site with grass and a loamy sand soil from a landscaped site with coniferous trees	CNT solutions added to the soil and incubated at 25 °C during 11 days	MWCNTs (specific surface area 237.1 m^2/g, specific volume 0.86 cm^3/g, diameter 15.1 ± 1.2, length 10–20 μm)	50; 500 and 5000 mg/kg	BET method, TGA, Raman, TEM	→ Enzyme activities showed a tendency to be repressed at medium CNT concentration. → Enzymatic activities and microbial biomass C and N were significantly lowered at high CNT concentration
[74]	Grassland soil from a natural reserve (sandy clay loam texture weakly acidic)	Soils incubated at room temperature for one year with CNT contamination	MWCNTs-1 (diameter of 23.3 ± 5.5 nm, specific surface area 72 m^2/g), MWCNTs-2 (diameter of 7.4 ± 1.9 nm, specific surface area of 500 m^2/g), MWCNTs-3 (diameter of 13.6 ± 4.6 nm, specific surface area of 200 m^2/g)	1000 mg/kg	SEM, TGA	→ The three types of CNTs reduced soil DNA and altered bacterial communities.
[72]	Sandy loam soil from a landscaped site dominated by grasses	Soils incubated with CNT powder and suspended forms of CNTs during 23 days	SWCNTs (average length of 1.02 μm, average diameter of 1.0 nm, purity > 90%, specific surface area of 1125.3 m^2/g), MWCNTs (specific surface area of 237.1 m^2/g)	30; 100; 300; 600 and 1000 mg/kg	TGA, BET method	→ SWCNTs significantly lowered activities of most enzymes and microbial biomass. → MWCNTs showed similar effects but at higher concentration.
[73]	Sandy loam soil from a grass dominated landscaped site	Soils incubated with CNT powder and suspended forms of CNTs during 25 days	SWCNTs (specific surface area of 1125.3 m^2/g, purity > 90%)	30; 100; 300; 600 and 1000 mg/kg	TGA, BET method	→ Biomass of major microbial groups showed a significant decrease with CNTs. → CNTs altered significantly microbial community composition.
[107]	Mix of soils (N.I)	Soils contaminated with CNT suspension	MWCNTs (diameter 25 nm, length of few microns)	50 mL of 50 or 200 mg/L of CNTs	TEM, RF-CVD	→ Bacteroidetes and Firmicutes were found to increase with CNT contamination. → Proteobacteria and Verrumicorbia decreased with increasing CNT concentration.
[118]	E.*coli*, P.*aeruginosa*, B. *subtilis and* S.*auresin* cultured in standard growth medium with and without CNTs	Bacteria incubated with CNTs overnight at 30 or 37 °C	SWCNTs (average diameter of 0.83 μm)	5; 10; 20; 40 and 80 mg/L	Raman spectroscopy, AFM, TEM, SEM and TGA	→ Individually dispersed CNTs were more toxic than aggregated CNTs. → Inhibiting cell growth and oxidative stress were not the major causes responsible for the death cells.
[75]	Sandy loam soil from a turf grass field	Soil samples exposed with CNTs during 14 days	SWCNTs (diameter from 0.9 to 1.44 nm)	250 and 500 mg/kg	Raman, TGA, SEM-EDX	→ Bacterial soil community was affected by CNT presence with major impact after 3 days of exposure but bacteria recovered completely after 14 days.
[76]	Soil from agricultural field with sand, silt and clay content of 12.9%, 76.1% and 11.0%	Soil incubated with CNTs during 60 days	SWCNTs (diameter <2 nm) and MWCNTs (diameter 10–20 nm)	0.2; 20 and 2000 mg/kg	–	→ SWCNTs at high concentration reduced mineralization. → MWCNTs at low concentration stimulated mineralization.
[119]	Sandy loam soil from a field site	Soils incubated with CNT suspension for 28 days	MWCNTs (diameter 30–50 nm, length 10–20 μm, purity > 95%)	10; 100; 1000 and 10,000 mg/kg	TEM, SEM, TGA	→ No effect on soil respiration, enzymatic activities and microbial community respiration at concentration lower than 10,000 mg/kg → At the highest treatment, abundance of some bacteria genera decreased
[120]	*Cupriavididus metallidurans* and *Escherichia coli*	Bacteria exposed to CNTs diluted in water at room temperature under gentle stirring during 24 h	MWCNTs (specific surface area 42 m^2/g, diameter 44 nm, length 1.5 μm)	10 and 100 mg/L	BET method, TEM CNT detection: TEM and STEM	→ CNTs accumulated on both bacterial strains

(*continued on next page*)

Table 1 (*continued*)

Article	Soils/bacteria	Culture conditions	CNT used[a]	Concentrations	CNT characterisation	Effects
[121]	Drummer soil (fine-silty) from continous corn no-till plots and a Tracy soil (coarse-loamy) from agricultural fields	Soils treated with CNTs weekly during six weeks	SWCNTs non functionalized, SWCNTs functionalized with polyethyleneglycol and SWCNTs functionalized with *m*-polyaminobenzene	60; 300 and 6000 mg/kg	–	→ Repeated application of SWCNTs can affect microbial activity and induce minor changes in soil metabolic activity → Functionalized CNTs seemed to be less toxic
[122]	*Paracoccus denitrificans*	Bacteria incubated in mineral medium contaminated by CNTs for 24 h	SWCNTs (average diameter of 1 −2 nm, length of 0.5−2 μm)	10 and 50 mg/L	SEM, TEM	→ CNTs were present outside and inside the bacteria. → The final nitrate concentration was higher with high concentration of CNTs → CNTs led to the transcriptional activation of the genes encoding ribonucleotide reductase in response to DNA damage and decreased the gene expressions genes involved in glucose metabolism and energy production

[a] All information provided on articles about CNT characterisation are given in this table.

organelle recycling in plant cells. According to the different studies that were identified, functionalized CNTs seem to enter more easily in plants compared to non-functionalized CNTs. It is important to precise that most of the studies on plants have been conducted in hydroponics conditions or only at the seed stage. Soil studies, more representative of environmental conditions, are a negligible part of the literature. However, no differences were found between CNT impacts in hydroponics or in soil.

Concerning impacts of CNTs on plants (Table 3): CNTs increased seed germination on a large range of concentrations (*i.e*: 40; 50; 100 and 500 mg/L) [21,88,89,103,104]. They can increase plant growth with a higher biomass production, higher flower production, or enhanced root elongation [86,89,91,94,96,103−109]. At the cellular level, CNTs were found to increase cell growth: MWCNTs enhanced growth of tobacco cell culture over a wide range of concentrations (0.005−0.5 mg/mL) [52,89].

On the other hand, in some studies CNTs were found to decrease plant growth: MWCNTs induced growth reduction and toxicity related to an increased generation of reactive oxygen species (ROS) in spinach at high concentration (125−1000 mg/L). They also caused necrotic lesions of leaf cells/tissues and changes of root and leaf morphology [102]. MWCNTs (10 mg/L) decreased cell dry weight, viability, chlorophyll content and superoxide dismutase (SOD) activity of *Arabidopsis thaliana* cell suspension [93]. SWCNTs had adverse effects on protoplasts and leaves through oxidative stress, leading to a certain amount of programmed cell death in *Arabidopsis thaliana* [110].

CNTs can also have no observed effect on plants, as reported in numerous studies. For example, Hamdi et al. [111] found no effect of MWCNTs functionalized and non-functionalized on seed germination of lettuce (*Lactuca sativa*). Lin and Xing [112] evidenced no effect of MWCNTs on seed germination and root length of several plants (radish, rape, ryegrass, lettuce, corn, and cucumber) like Larue et al. [20] in wheat and rapeseed.

Looking at the gene level, CNTs seem to up-regulate genes involved in cell division/cell wall formation in tomato plants [114]. SWCNTs promoted rice root growth through the regulation of expression of the root growth related genes (*Nt*LRX1 and CyCB) [115]. MWCNTs were also observed stimulating the expression of water channel genes (aquaporins) [82,83,98,107]. Aquaporins are central components in water-plant relationships, as they are essential for root water uptake, seed germination, cell elongation, reproduction and photosynthesis [114]. The overexpression of aquaporin genes can contribute to cell growth leading to overall plant growth. They also up-regulate genes involved in response to pathogens meaning that CNTs could be sensed by plants as a stress similar to herbivore attack [61]. Other authors also found that CNTs provoke repression of pathogen-activated genes and salicylic acid-mediated pathways in *Arabidopsis thaliana* [116]. Same authors demonstrated that there is a greater similarity in the plant response to nanoparticles of different chemical nature, than there is with other environmental stress (salinity, biotic stress …).

CNT impacts on plants can be different according to the types of CNT used (functionalized or not, number of walls) as shown in Fig. 4. Phytotoxicity varied between CNTs non-covalently functionalized with poly-3-aminobenzenesulfonic acid (PABS) and non-functionalized CNTs [113]; the first ones affecting more root length than the second ones. In another study, non-functionalized CNTs inhibited root elongation in tomato but enhanced it in onion and cucumber while functionalized CNTs inhibited root elongation in lettuce [113]. Toxicity of MWCNTs can increase sharply as the diameter of the agglomerates decreased [93], suggesting that a better dispersion could enhance the toxicity. Functionalized CNTs are usually better dispersed but the literature does not always describe them as more toxic, this point is still in debate. Moreover,

Table 2
Studies on CNT behaviour and impacts on soil macroorganisms.

Reference	Species	Growth conditions	CNT used[a]	Concentrations	CNT characterisation	Effects
[82]	Earthworms (*Eisenia fetida*)	Earthworms incubated in soil contaminated by CNTs during 14 days	MWCNTs	30 and 300 mg/g	–	→ Immune cells morphometric alterations, lysosomal membrane destabilization, acetylcholinestrase inhibition and metallothionein tissue concentration changes are highly sensitive to MWCNTs.
[84]	Eartworms (*Caenorhabditis elegans*)	Earthworms incubated on nematode growth medium with and without CNTs during 72 h	MWCNTs	500 mg/L	–	→ Phagocytosis could be a potential mechanism of uptake of CNTs and oxidative stress a potential mechanism of toxicity.
[81]	Earthworms (*Eisenia fetida*)	Earthworms grown on artificial soil contaminated with CNTs during 7 days	MWCNTs (diameter of 10 nm and length 9–20 μm, specific surface area of 500 m^2/g)	100 and 1000 mg/kg of dry soil	BET method, TEM	→ MWCNTs absorbed nonylphenol caused much more adverse effects to the earthworms than each chemical alone.
[123]	Earthworms (*Eisenia fetida*)	Earthworms grown on artificial soil contaminated with CNTs during 14 days	MWCNTs (purity >99.5%, average inner diameter of 10 nm and length of 10 μm, specific surface area of 500 m^2/g)	50; 500 and 1000 mg/kg of dry soil	BET method, TEM, X-ray diffraction, Raman	→ No mortality was found in soil contaminated with CNTs even at the highest concentration. → DNA damages were found in earthworms at relatively low concentration of CNTs in the medium.
[83]	Earthworms (*Eisenia fetida*)	Earthworms grown in sandy loam soil spiked with CNTs during 28 days	MWCNTs (diameter from 30 to 50 nm, length from 10 to 20 μm, purity >95%)	3000 mg/kg of soil	TEM	→ Low bioaccumulation factor of CNTs in earthworms.
[77]	Earthworms (*Eisenia fetida*)	Soils from field sites were spiked with CNTs, earthworms were added and stayed for 14 days	^{14}C-MWCNTs (diameter from 30 to 70 nm, purity > 99%),^{14}C-SWCNTs (diameter from 1 to 2 nm, purity > 91%)	30 and 300 mg/kg	TEM, TGA Raman	→ Adsorption of CNTs on the tissues of earthworms was minimal.
[78]	Earthworms (*Eisenia fetida*)	Earthworms grown in soil spiked with CNTs and pyrene for 28 days	MWCNTs (purity 99%, diameter 30–70 nm), SWCNTs (purity 91%, diameter 1–2 nm)	30 and 300 mg/kg	TGA, Raman, TEM	→ Both CNTs at the highest concentration decreased pyrene bioaccumulation. → Presence of CNTs enhanced pyrene elimination rates.
[79]	Earthworms (*Eisenia veneta*)	Earthworms cultured three types of soils (organic carbon fractions 5.7%, 1.6% and 3.9%) contaminated by CNTs during 28 days	^{14}C-MWCNTs (diameter between 30 and 70 nm)	500 mg/kg of dry soil	TEM, SEM, TGA, electrophoretic mobilities (Malvern Zetasizer Nano ZS)	→ Limited absorption of CNTs into organisms tissues. → Earthworms can easily eliminate accumulated CNTs.
[80]	Earthworms (*Eisenia veneta*)	Earthworms in loamy sand soil were fed with foods contaminated by CNTs during 21 days	DWCNTs (diameter of 10–30 nm, length of 5–15 μm, specific surface area of 1,255,637 nm^2, purity of 99.5%)	50; 100; 300 and 495 mg/kg of dry food	–	→ Reproduction of the studied earthworms was affected by CNTs → The most sensitive toxicological parameter was reproduction (cocoon production), with no effect on hatchability, survival or mortality
[85]	Earthworms (*Eisenia fetida*)	Earthworms on artificial soil contaminated by CNTs during 14 days	MWCNTs (purity > 95%, average length of 10 μm, specific surface area of 500 m^2/g)	1000 mg/kg of dry soil	TGA, TEM, BET method	→ CNTs induced slight toxicity compared to sodium pentachlorophenate → Expression of enzymatic biomarkers was different with PCP-Na and CNTs at the same time than PCP-Na or CNTs alone

Abbreviations: CBNMs: Carbon Based Nanomaterials; UV–vis–NIR: Ultraviolet–Visible and Near-Infrared Spectroscopy; TEM: Transmission Electron Microscopy; AFM: Atomic-Force Microscopy; SEM: Scanning Electron Microscopy; GC-MS: Gas Chromatography Mass Spectrometry; GC-ECD: Gas Chromatography with Electron Capture Detector; EDS: Energy-Dispersive X-ray Spectroscopy; TGA: Thermo-Gravimetric Analyse; BET: Brunauer-Emmett-Teller; QPCR: Real-Time PCR; Integrated PC/PT scanning cytometry: Integrated PhotoThermal and PhotoAcoustic scanning cytometry; ICP MS: Inductively Coupled Plasma Mass Spectrometry; FTIR: Fourier Transform Infrared Spectroscopy; EDX: Energy-dispersive X-ray spectroscopy; N.I: Non Informed.

[a] All information provided on articles about CNT characterisation are given in this table.

Table 3
Studies on CNT behaviour and impacts on plants.

Reference	Plant used	Culture conditions	CNT used[a]	Concentrations	Detection and characterisation techniques	Effects
[52]	Tomato *(Solanum lycopersicum cv Micro-Tom)*	Seeds germinated on agar medium with and without CNTs for 2 months	SWCNTs functionalized with QDs and non-functionalized	50 mg/L	CNT characterisation: UV−vis−NIR, TEM CNT detection: Raman, UV light radiation	→ Addition of QDs to CNTs dramatically changed the biological variability by accelerating leaf senescence and inhibiting root formation. → CNTs only induced "positive" effects (increase of the chlorophyll content and total weight of the root system).
[86]	Cabbage (Brassica oleacera)	One-week-old germinated seed grown in medium with and without CNTs and with and without NaCl	MWCNTs (diameter 6−9 nm, length 5 μm, purity 95%)	10; 20; 40 and 60 mg/L	CNT characterisation: TEM	→ CNTs entered in cells with higher accumulation under salt stress → CNTs had positive effect on growth in NaCl-treated plants. → CNTs induced changed in the lipid composition, rigidity and permeability of the root plasma membranes relative to salt stressed plants. → CNTs enhanced aquaporin transduction.
[95]	Spinach (*Amaranthus tricolor*)	Seeds immersed in CNTs suspension for one night and placed in filter paper until germination, then transferred to plastic pots for hydroponic culture with and without CNTs for 15 days	MWCNTs (diameter around 11 nm, length < 1 μm)	125; 250; 500 and 1000 mg/L	CNT characterisation: AFM, SEM, TEM CNT detection: Raman, SEM, TEM	→ CNTs induced growth reduction and toxicity due to the ROS. → CNTs caused necrotic lesions of leaf cells/tissues and changed of root and leaf morphology. → CNTs were found in leaves.
[102]	Lettuce (Lactuca sativa), rice (*Oryza sativa*), cucumber (*Cucumis sativus*), red spinach (*Amaranthus tricolor*), lady's finger (*Abelmoschus esculentus*), chili (*Capsicum anuum*), soybean (*Glycine max*)	Seedlings transferred in medium with and without CNTs and growth for 15 days	MWCNTs (diameter around 13 nm, length around 1 μm)	20; 200; 1000 and 2000 mg/L	CNT characterisation: SEM, TEM	→ CNTs reduced root and shoot length. → CNTs increased cell death and electrolyte leakage. → Very little or no toxic effects were found for chili, lady's finger and soybean. → Red spinach and lettuce were more sensitive to CNTs.
[112]	Cabbage (*Brassica oleracea*), carrot *(Daucus carota)*, cucumber (*Cucumis sativus*), onion (*Allium cepal*), tomato (*Lycopersicon esculentum*) and lettuce (*Lactuca sativa*)	Seeds exposed to CNTs during 24 and 48 h	SWCNTs functionalized and non-functionalized (diameter 8 nm, length of few microns)	28; 160; 900 and 5000 mg/L	CNT characterisation: SEM	→ CNTs and fCNTs inhibit root elongation of four crop species (cucumber, inion, lettuce and tomato). → Phytotoxicity varied between CNTs and fCNTs, with CNTs affecting more species. → Tomato was the most sensitive species. → Microscopy images showed the presence of NTCs on the root surface.
[98]	Corn (*Zea mays*)	Germinated seeds cultivated in soil with and without CNTs for 40 days	OH-functionalized SWCNTs, COOH-functionalized SWCNTs and non-functionalized SWCNTs (diameter 1−4 nm, length 5−30 μm, purity >90% wt%)	10 and 100 mg/kg (wt/dry wt)	CNT characterisation: TEM, microwave induced heating method	→ CNTs accumulated mostly in roots, with minimal accumulation in stems and leaves.
[70]	Zucchini (Cucurbita pepo cv Costata Romanesco), tomato (*Solanum lycopersicum*), soybean (*Glycine max*), corn (*Zea mays*)	3 to 7 day-old seedlings (depending of the species) grown in soil contaminated with CNTs and pesticides during 28 days	MWCNTs (95% purity, diameter 13−18 nm, length 10−30 μm)	500; 1000 and 5000 mg/kg	CNT characterisation: GC-MS and GS-ECD	→ CNTs suppressed in a dose-dependent fashion the bioaccumulation of weathered chlordane and DDx. → CNTs were found in root and shoot tissues.

[124]	Cabbage (*Brassica oleracea*)	Seedlings were grown in nutrient solution or in soil with carbamazepine and CNTs	Pristine CNTs and carboxyl-functionalized CNTs (purity 95%, diameter <8 nm, length 10−30 μm, specific surface area 500 m^2/g)	50 mg/L (hydroponic experiments), 50 mg/kg (soil experiments)	–	→ Biomass enhancement was observed on plants grown with CNTs. → Co-exposure with CNTs suppressed carbamazepine accumulation. → Functionalized CNTs enhanced carbamazepine translocation potential.
[125]	Lettuce (*Lactuca sativa*)	Seeds germinated on medium contaminated with and without CNTs, with and without humic acid, after 10 days pesticides were added, growth for 19 days in total	MWCNTs functionalized (diameter <8 nm, length 10 −30 μm, purity 95%), MWCNTs non-functionalized (diameter 13 −18 nm, length 3−30 μm, purity > 99%)	1000 mg/L	–	→ CNTs did not influence seed germination. → CNT presence and type significantly influenced pesticide availability.
[87]	Rice (*Oryza sativa*)	5-day-old seedlings tranplanted in tubes with nutrient solution with CNTs during 15 days	Hollow MWCNTs, Fe-filled CNTs, Fe-Co-filled CNTs (typical diameters of dozens of nm)	0; 10; 50 and 300 mg/L	CNT characterisation: TEM, EDS	→ The three types of CNTs had toxic effects on rice seedlings, and inhibited the growth and development of roots and shoots. → The C:N ratio in rice roots significantly increased after treatments with CNTs, and all three types of CNTs had the same effect. → CNTs penetrate cell wall and cell membrane, they could be transported to shoots.
[126]	Henbane *(Hyoscyamus niger)*	Seeds exposed to different concentrations of CNTs during 14 days under drought stress	SWCNTs (outer and inner diameter of 1−3 and 0.9 −2 nm and length of 5 −30 μm)	50−800 mg/L	CNT characterisation: TEM, SEM, Raman, TGA, BET and X-ray diffraction	→ SWCNTs at low concentrations induced tolerance in seedlings against low to moderate level of drought by enhancing water uptake and activating plant defense system.
[88]	Tomato (Solanum lycopersicum cv Micro-Tom)	Seeds placed on MS medium without or with CNTs for 3, 12 and 20 days	MWCNTs (purity higher than 98%)	10; 20 and 40 mg/L	CNT characterisation: SEM, TEM, TGA, Raman CNT detection: TEM	→ MWCNTs can penetrate thick seed coat and support water uptake inside cells. → Positive effects of MWCNTs on seed germination.
[61]	Tomato (Solanum lycopersicum cv Micro-Tom)	Seeds exposed to CNTs during 10 days	MWCNTs functionalized	50 mg/L	CNT characterisation: TEM CNT detection: microaray analysis, real time QPCR, integrated PA/PT scanning cytometry, Raman	→ MWCNTs induce previously unknown changes in gene expression in tomato leaves and roots, particularly, up-regulation of the stress-related genes. → Detection of MWCNTs in roots, leaves, and fruits down to the single nanoparticle and cell level.
[114]	Tobacco cells *(Nicotiana tabacum* cv Havana*)*	Cells grown on MS medium without and with CNTs for 30 days	MWCNTs (diameter 20 nm, length from 500 nm to 1 μm)	0.1; 5; 100 and 500 mg/L	CNT characterisation: TEM and Raman	→ Enhance the growth of tobacco cell culture in a wide range of concentrations (5−500 μg/mL). → Correlation between the activation of cell growth exposed to MWCNTs and the upregulation of genes involved in cell division/cell wall formation and water transport.
[107]	Tomato (Solanum lycopersicum cv Micro-Tom)	Plants grown in soil supplemented with CNTs during 10 days	MWCNTs (diameter 25 nm, length of few microns)	50 and 200 mg/L	CNT characterisation: TEM CNT detection: TEM, Raman	→ Plants grown in soil supplemented with CNTs produce two times more flowers and fruits compared to plants grown in control soil.
[90]	Barley hybrid Robust (*Hordeum vulgare*), corn hybrid N79Z 300 GT (*Zea mays*) and soybean hybrid S42-T4 (*Glycine max*)	CNTs deposited on seed surface by airspray techniques or added in growth medium of seeds, 10 days of exposure	MWCNTs functionalized (diameter from 15 to 40 nm, length of several μm)	50; 100 and 200 mg/L	CNT characterisation: TEM and Raman	→ MWCNTs for both deposit technics penetrate seed coats of all tested species and activate germination of MWCNT-exposed seeds. → Application of CNTs to the seeds of the three studied species can stimulate expression of water channel genes (aquaporins).
[104]	Soybean hybrid S42-T4 (*Glycine maxl)*, barley hybrid Robust (*Hordeum vulgare*), corn hybrid N79Z 300 GT (*Zea mays*), tomato cv Micro-Tom (*Solanum lycopersicum*), switch grass (*Panicum virgatum*), rice cv. *Cypress (Oryza sativa)*, tobacco cell culture (*Nicotiana tabacum*)	Seeds germinated on medium contaminated with and without CNTs for 10 days (corn), 11 days (barley and soybean), 12 days (rice) and 20 days (tomato and switch grass)	SWCNHs (nanohorns)	25; 50 and 100 mg/L	CNT characterisation: SEM, TEM, TGA, Raman CNT detection: TEM, microwave induced heating technique	→ CNHs activated seed germination and enhanced growth of different organs of corn, tomato, rice and soybean. → CNHs increased growth of tobacco cells. → CNHs were found inside cells → CNHs affected expression of a number of tomato genes involved in stress responses, cellular responses and metabolic processes.

(*continued on next page*)

Table 3 (*continued*)

Reference	Plant used	Culture conditions	CNT used[a]	Concentrations	Detection and characterisation techniques	Effects
[89]	Tomato cv Micro-Tom (*Solanum lycopersicum*), tobacco callus cells (*Nicotiana tabacum)*	Callus cells exposed to growth medium with and without CNTs, seeds grown in medium without and with CNTs	COOH-functionalized MWCNTs (diameter 13 −18 nm, length 1−12 μm), COOH-functionalized MWCNTs (diameter < 7 nm, length 0.5−2 μm), helical MWCNTs (diameter 100 −200 nm, length 1−10 μm)	50 and 100 mg/L	CNT characterisation: TEM, Raman	→ CNTs activated cell growth, germination and plant growth. → CNTs were found inside seeds. → Helical CNTs affected a number of genes involved in cellular and metabolic processes and response to stress factors. → CNTs upregulated expression of the tomato water channel gene.
[20]	Wheat (*Triticum aestivum*), rapeseed (*Brassica napus*)	15-day-old seeds in CNTs suspension for 7 days	MWCNTs (diameter 41.2 nm, specific area 42 ± 2 m^2/g)	100 mg/L dispersed with arabic gum or humic acid	CNT characterisation: TEM CNT detection: TEM, Raman	→ Less than 0.005‰ of the applied CNT dose was taken up by plant roots and translocated to the leaves. → This accumulation does not impact plant development and physiology. It does not induce any modification in photosynthetic activity or cause oxidative stress in plant leaves.
[112]	Rape (*Brassica napus*), radish (*Raphanus sativus*), ryegrass (*Lolium perenne*), lettuce (*Lactuca sativa*), corn (*Zea mays*) and cucumber (*Cucumis sativus*)	Seeds exposed to CNTs during 5 days	MWCNTs (diameter 10 −20 nm, length 1−2 μm, purity > 95%, surface area 126 m^2/g)	20; 200; 2000 mg/L	CNT characterisation: BET	→ CNTs did not impact seed germination and root length.
[93]	Thale cress T87 suspension cells *(Arabidopsis thaliana)*	72-h-old cell cultivation exposed to CNTs in the cell suspension Cells exposed to CNTs for 2, 3, 4, 5, 6 and 7 days	MWCNTs (average diameter 9.5 nm, average length 1.5 μm, surface area 250−300 m^2/g)	10; 60; 100 and 600 mg/L	CNT characterisation: TEM, BET, ICP-MS CNT detection: TEM	→ CNTs decreased cell dry weights, cell viabilities, cell chlorophyll contents and superoxide dismutase activities. → Toxicity of CNTs increased sharply as the diameters of the agglomerates of MWCNTs become smaller.
[86]	Cabbage (*Brassica oleacera*)	5-day-old seeds placed in containers with continuously-aerated Hoagland nutrient, exposure for 7 days	MWCNTs (diameter between 6 and 9 nm, length of 0.1−0.5 μm)	Exp 1: 10; 20; 40 and 60 mg/L Exp 2: 10 mg/L with NaCl	CNT characterisation: TEM	→ "Positive" effect on the growth under both saline and non-saline conditions. → Increase Na concentrations in roots of Na-Cl treated plants.
[94]	Alfalfa (*Medicago sativa*), wheat (*Triticum aestivum*)	Seeds cultivated in medium contaminated by CNTs during 6 days	MWCNTs functionalized with Fe3O4 (67.2% purity, 10.9 ± 1.9 nm, 116.1 m^2/g)	40; 80; 160; 320; 640; 1280; 2560 mg/L	CNT characterisation: TGA, TEM, Raman, N2 adsorption/desorption isotherms CNT detection: TEM, Raman	→ CNTs did not impact germination of both species. → CNTs enhanced root elongation. → CNTs were absorbed onto the root surfaces without significant uptake or translocation.
[127]	Red clover (*Trifolium pratense*)	5-day-old seeds transferred in agricultural soil (brownearth with a sandy loamy to loamy fine fraction) with and without CNTs and growth during 14 weeks	MWCNTs (diameter 20 −30 nm, length 10−30 μm, purity >95%)	10; 100 and 1000 mg/kg	–	→ CNTs did not affect plant biomass and arbuscular mycorrhizal fungi root colonization. → CNTs decreased the number of flowers → CNTs increased nitrogen fixation.
[53]	Mustard (*Brassica juncea*)	Seeds germinated on petri dishes with and without CNT suspension until complete germination (radicle attained a length of 1 mm)	MWCNTs (purity >60%, diameter around 30 nm)	2.3; 6.9; 23 and 46 mg/mL	CNT characterisation: SEM, FTIR, X-ray diffraction CNT detection: FTIR, SEM	→ CNTs increased moisture content of seeds and enhance water absorption machinery of root tissues. → CNTs can be transported through the plant vascular cylinder.
[103]	Cress (*Lepidum sativum*), sorgo (*Sorghum saccharatum*), tomato (*Solanum lycopersicon*), radish (*Raphanus sativus*), cucumber (*Cucumis sativus*)	Seeds germinated on four different sewage sludges spiked with CNTs with a storage during 7 and 31 days for aging	MWCNTs (diameter <10 nm, surface area 357 m^2/g, purity >95%), MWCNTs (diameter 40 −60 nm, surface area 73 m^2/g, purity >95%)	0.1; 1 and 5 g/kg	–	→ CNT influence on sludge toxicity varied with respect to CNTs' outer diameter, type of sewage sludge and plants tested. → CNTs had positive effects on seed germination and root growth of two sewage sludge.

[128]	Carrot (*Daucus carota*)	Seeds exposed to CNTs during 5 days in petri-dishes with and without CNTs and AgNPs during 5 days	MWCNTs (median diameter 6.6 nm, length of 5 μm)	10; 100; 200; 500; 1000 and 2000 mg/L	–	→ CNTs did not significantly affect seed germination and seedling growth. → CNTs decreased H_2O_2 levels → CNTs reduced levels of a seed protein, DcHsp17.7, during seed germination and increased chlorophyll content.
[21]	Tomato (Solanum lycopersicum cv Micro-Tom)	Seeds sonicated in suspension with and without CNTs, and seeds germinated in petri dishes until germination (until root (radicle) is visible)	CBNMs (hydrophilic fullerols and hydrophobic MWNTs)	50 mg/L with gallic acid	CNT characterisation: TEM, SEM and microRaman	→ CNTs did not penetrate seed coat. → CNTs enhanced germination and seedling length and weight.
[99]	Periwinkle (*Caranthus roseus*)	Perwinkle cell suspension culture incubated with CNT suspension during 3 h at 25 °C or 4 °C	MWCNTs (purity 95%, average outer diameter 20−30 nm, length 0.5−2 μm)	10; 20; 40; 60 and 80 mg/L	CNT detection: TEM, confocal microscopy imaging	→ CNTs are entering passively through the cell membrane and it's not associated with the endosmal route. → Isolated CNTs were observed inside cells as a result of a direct penetration of the plasma membrane. → No CNTs found in any organelles associated with endocytosis cycle. → CNT distribution followed a size distribution of short CNTs (30−100 nm) inside organelles, while long CNTs (>200 nm) were found inside subcellular structures.
[110]	Thale cress (*Arabidopsis thaliana* ecotype Columbia Col-0)	Incubation of Thale cress cells with suspension of CNTs	Cup-staked CNTs (average outer diameter 60−100 nm, length 1−100 μm)	–	CNT detection: confocal microscopy imaging, AFM	→ CNTs participate in cell biochemical reactions. → CNTs were detected into the structure of tracheids and showed such mutual and parallel arrangement with a lignin polymer.
[34]	Thale cress (*Arabidopsis thaliana)*	Incubation of Thale cress cells with CNTs for 3 h	Cellulase-immobilized cup-staked CNTs	–	CNT characterisation: AFM, CNT detection: AFM, epifluorescence microscopy	→ Cellulase-imobilized CNTs penetrated the thick cellulosic cell wall and they are transported into the cell.
[129]	Mung bean (*Phaseolus radiatus)* and Radish (*Raphanus sativus*)	Seeds germinated on sediments (organic carbon content 1.58%, 47.6% of clay, 28.87% of silt and 23.53% of sand) spike with CNTs, phenanthrene and cadmium during 72 h	ThreeMWCNTs (purity>95%) with different outer diameter (10−20 nm, 30−50 nm, >50−98 nm) and specific surface area of respectively 134 $m^2\ g^{-1}$, 103 m^2, 206 g^{-1}, and 70.1 $m^2\ g^{-1}$	0.5%, 1.0%, or 1.5% (w/w)	CNT characterisation: SEM, FTIR, and BET method	→ MWCNTs showed a better adsorption performance with phenanthrene and cadmium (II) compared with sediments. → MWCNTs did not inhibit significantly the germination but root growth was more sensitive than biomass production to the changes of contaminant concentration.
[110]	Thale cress *(Arabidopsis thaliana)*, rice (*Oryza sativa* subsp. *Japonica* cv. Nipponbare)	Protoplasts cultured in CNTs Injection of CNTs into intact leaves	SWCNTs (diameter 1−2 nm, length 5−30 μm, purity 90%)	5; 25; 100 and 250 mg/L	CNT characterisation: Fluorescence, TEM	→ CNTs had adverse effects on protoplasts and leaves through oxidative stress, leading to a certain amount of programmed cell death.
[96]	Sainfoin (*Onobrychis arenaria*)	Seeds germinated on petri-dishes with and without Taunit suspension (CNTs) for 10 days	Taunit suspension: loose black powder composed of grainy agglomerates containing MWCNTs (diameter 5−10 nm, length of at least 2 μm, purity 98%)	100 and 1000 mg/L	CNT characterisation: TEM and light electron microscopy	→ CNTs stimulated the growth of roots and stems, and enhanced the peroxidase activity in these part of plants → CNTs were found in leaves and stems tissues.
[130]	Zucchini (*Cucurbita pepo* cv Costata Romanesco)	Seeds exposed to CNTs for 5 and 12 days 4-day-old seeds in CNT suspension during 15 days	MWCNTs (purity >99%, number of walls from 3 to 15)	1000 mg/L	–	→ CNTs did not impact seed germination and root length. → CNTs reduced biomass of plants of the 15 day hydroponic trial.
[109]	Date palm (*Phoenix dactylifera*)	8-month-old callus cells subcultured four times with 6 weeks intervals on CNT media	MWCNTs (diameter 11−170 nm, length 5−9 μm)	0.05; 0.1 mg/L	CNT characterisation: TEM	→ Low concentrations of CNTs promoted callus fresh weight, increased number of germinated embryos, shoot length and leaf number and enhanced root number, root length, plantlet length and hairy roots. → CNTs can penetrate plant tissues and enter its cells → CNTs can facilitate the adsorption or transportation of nutrients into plant tissues.

(*continued on next page*)

Table 3 (*continued*)

Reference	Plant used	Culture conditions	CNT used[a]	Concentrations	Detection and characterisation techniques	Effects
[131]	Rice (Oryza sativa L.)	Callus cells of three-month-old plants transferred to a cell suspension culture, after 10 days add of CNTs for 4 days	MWCNTs (diameter 20 −40 nm, length 0.5−50 μm, surface area 3.14×10^{-2} - 6.28 μm^2)	0.05 and 0.1 g/L	CNT characterisation: SEM	→ CNTs decreased cell density, possibly indicating a self-defense response. → CNTs interacted directly with rice cells and may had a detrimental effect on rice growth.
[132]	Rice (Oryza sativa L.)	6-day-old cell culture exposed to CNTs during 6 days	MWCNTs (diameter 10 −30 nm, length 5−15 μm, surface area 86 m^2/g, purity 95%)	20 mg/L	CNT detection: TEM	→ CNTs increased ROS content and decreased cell viability. → Individual tubes found in contact with cell walls.
[91]	Corn (*Zea mays*)	Seeds germinated on medium with and without CNTs for 7 days	MWCNTs (diameter 6 −9 nm, length 5 μm, purity >95%)	20 mg/L	CNT characterisation: SEM	→ CNTs enhanced germinative growth at low concentration but depressed it at higher concentration. → CNTs improved water absorption, plant biomass and concentration of the essential Ca, Fe nutrients. → CNTs perforated the black-layer seed-coat while in presence of $FeCl_2$/$FeCl_3$ they didn't perfortate.
[105]	Common gram (*Cicer arietinum*)	One-day-old seeds exposed to CNTs	MWCNTs (diameter 10 −30 nm)	6 mg/L	CNT characterisation: EDX, TEM, Raman CNT detection: SEM, TEM, fluorescence	→CNTs increased growth rate of roots, shoots and branching. → CNTs enhanced water absorption.
[106]	Tomato (Solanum lycopersicum cv Micro-Tom)	Seeds cultivated in medium contaminated by CNTs during 28 days	MWCNTs (diameter 8 −35 nm, several micrometers in length, 94% purity), MWCNTs purified by HCl washing and sonification (98% purity), MWCNTs further oxidized and decorated with carboxylic groups, MWCNTs sonicated in acetone, MWCNTs coated with PEG	40 mg/L	CNT characterisation: TEM, SEM, TGA, Raman, zeta potential	→ Highest increase in plant growth was observed for plants exposed to well dispersed MWCNTs and MWCNTs functionalized with strong negative groups. → Production of tomato water channel protein was activated in plants exposed to MWCNTs functionalized with various groups.
[133]	Soybean (*Glycine max)*	Seeds cultivated in petri dishes with filter paper contaminated by CNTs during 10 days; seeds cultivated in CNTs suspension during 36 h	MWCNTs (purity above 98%, outer diameter of 20 −70 nm, inner diameter of 5−10 nm and length of >2 μm)	1000 mg/L	–	→ MWCNTs induced oxidative stress in radicle tips which coincided with MWCNTs accumulation. → MWCNTs reduced Zn translocation from the cotyledons to the seedlings. → MWCNTs exhibited adsorption potential for Zn and Cu.
[134]	Thale cress *(Arabidopsis thaliana)*, soybean (*Glycine max*), rice (*Oryza sativa*), maize (*Zea mays*)	Plants in hydroponic conditions with semisolid medium (MS basal medium with vitamins and sucrose) CNTs and/or SPAOMs during 25 days	14C MWCNTs (specific surface area of 111 m^2/g, specific radioactivity of 0.1 mCi/g, surface oxygen content of 8.6%, diameter 36.5 ± 12,7 nm, length 350 nm)	0.45; 0.9; 2.25 and 4.5 mg/L	CNT characterisation: X-ray photoelectron spectroscopy, thermal gravimetric, SEM CNT detection: liquid scintillation counting	→ Changes in biochemical parameters were much more sensitive than physiological parameters. → CNTs could alleviate the toxicity of SPOAMs to *Arabidopsis.* → Hydrodynamic diameter did not significantly affect CNTs uptake
[97]	Corn *(Zea mays*), soybean (*Glycine max*)	7-day-old germinated seeds were added in medium contaminated with and without CNTs during 18 days in hydroponic conditions	pristine-MWCNTs, amine (NH2)-functionalized MWCNTs, carboxylate (COOH)-functionalized MWCNTs (diameter 20 −30 nm, length 0.05 −2.0 μm)	10; 20 and 50 mg/L	CNT characterisation: TEM	→ The three types of CNTs were directly taken up and translocated to roots, stems and leaves → CNTs accumulated in phloem and xylem cells within specific intracellular sites like the cytoplasm, cell wall, cell membrane, chloroplast and mitochondria → CNTs stimulated maize growth and inhibited soybean growth

[135]	Rice (*Oryza sativa*)	Germination of seeds during 7 days in Hoagland medium with and without CNTs, then transplanted to the normal Hoagland medium	SWCNTs (diameter 1–2 nm, length around 30 μm), MWCNTs (diameter 20 –40 nm, length 10–30 μm)	5; 20 mg/L	CNT characterisation: TEM	→ SWCNTs located in the intercellular space while MWCNTs penetrated cell walls in roots → CNTs promoted rice root growth through the regulation of expression of the root growth related genes → CNTs caused a similar histone acetylation and methylation statuses across the local promoter region of the Cullin-RING ligases 1 (CRL1) gene and increased micrococcal nuclease accessibility of this region, which enhanced this gene expression.
[136]	Corn (*Zea mays*)	Plants cultivated in soil contaminated with CNTs, pyrene and methypyrene during 26 days	MWCNTs (inner and outer diameter of the MW ranged in 5–15 nm and 50–80 nm, respectively, withthe length ranging in 10 –20 mm)	50 and 3000 mg/kg	–	→ Concentrations of both pyrene and 1-CH_3-pyrene decreased with increasing amendment level of MWCNTs, indicating an increasing suppression their bioaccumulation and translocation in plants.

[a] All information provided on articles about CNT characterisation are given in this table.

functionalisation of CNTs induced strong treatments which are reducing the CNT length. It is thus difficult to determine if functionnalisation or CNT matters most for the toxicity. Serag et al. [99] reports that MWCNTs larger than 200 nm accumulated in subcellular organelles while shortest ones (30–100 nm) were found into vacuoles, nucleus and plastids. However, it is the only paper that report this difference between short and long MWCNTs. It is not possible to compare the effect of CNT length in between different papers because experiment conditions and CNTs were different.

Controversial effects of CNTs have been evidenced in plants. It is important to standardize evaluation methods to better understand the results and to allow a better comparison between studies.

2. Conclusion

CNTs represent a large group of carbon-based nanomaterials which can differ in many ways such as diameter, length, number of layers, impurities or surface modification. In the literature, a variety of different CNTs have been used, with different suspension media and various suspension protocols. Despite the large range of CNTs, general conclusions about behaviour and impacts of CNTs on the terrestrial ecosystem can be drawn from the reviewed studies. First, changes in surface properties or adsorption of other compounds (cocktail effect) determine CNT environmental behaviour. Indeed, non-functionalized CNTs are hydrophobic, and thus difficult to disperse, they agglomerate rapidly. Functionalisation of CNTs makes them more hydrophilic. CNTs have strong adsorption properties, which can be used intentionally in remediation applications to remove pollutants but may also lead to the binding of compounds present in the environment such as natural organic matter or contaminants with a Trojan horse effect. In general, CNTs will remain in soil and will not reach aquifers. Soil macroorganisms, and earthworms in particular, have a low bioaccumulation of CNTs due to an efficient depuration system. In plants, CNTs seem to penetrate in both seeds and roots and are subsequently translocated into the upper part of plants to edible parts. Very low concentrations were found in plants.

CNT impacts on terrestrial ecosystem are divided in 3 categories. Some studies agreed that CNTs can increase plant growth and soil microbial activity but also the development of soil macroorganisms. Other studies reported opposite effects. Finally, a number of other studies concluded that CNTs had no influence. Obviously, CNT toxicity varied according to their intrinsic characteristics, the medium type and the dispersion method. CNTs could be perceived as an environmental stress. Organisms will react differently to defend themselves against this stress, for example by the overexpression of some genes. This could contribute to cell growth and in turn to organism growth. The impact that one could qualify as “positive” due to the growth increase may be a simple stress response to an environmental factor but further investigations in more environmentally relevant conditions would be needed to conclude. There is a gap between the high concentration range tested on organisms in the literature so far and the prediction of expected concentration of CNTs in soils. There is a lack of studies on CNT impacts and at realistic concentrations.

Detection of CNTs in carbonaceous matrices still constitutes one of the main challenges of this field of research. The development of quantitative techniques for accurate measurement of CNTs in biological and environmental samples will help a lot understanding the transfer of CNTs, their fate and impact in complex soil-based ecosystems.

There is also a lack of standardized methods, leading to controversial results on CNT impacts, making difficult the comparison and analysis of earlier works. It is important to make the connection between exposure conditions and effect, this will help

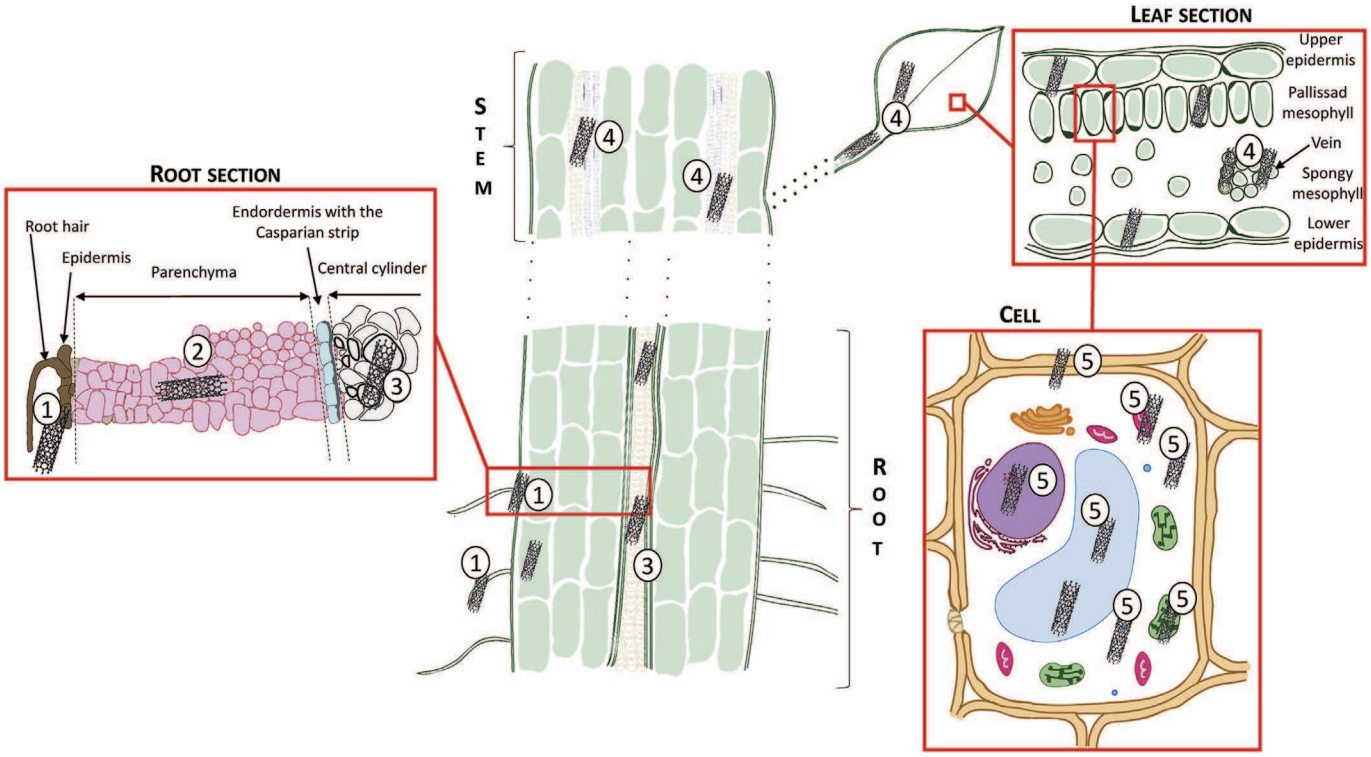

Fig. 3. Uptake and distribution of carbon nanotubes on plants [20,61,70,87,93,94,96–102]. CNTs are not at the good scale. In the cell, in blue it is the vacuole, in green the chloroplasts, in purple the nucleus with the grainy endoplasmic reticulum, in orange the smooth endoplasmic reticulum, and in blue the plasmode.

1. CNTs can enter plant roots through osmostic pressures, capillary forces, pores on cell walls, intercellular plasmadesmata or through direct penetration.
2. CNTs traverse through both cell wall and cell membrane through endocytosis.
3. CNTs may share the vascular system with water and nutrients and may be transported via transpiration through the upper part of plants.
4. CNTs are found in the upper part of plants. In leaves, they are mainly found in the leaf xylem.
5. In cell, CNTs accumulate mainly in cytoplasm, cell wall, cell membrane, chloroplast, mitochondria and plasmodes.

(A colour version of this figure can be viewed online.)

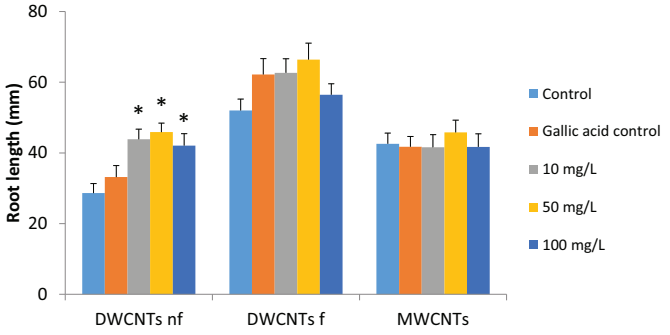

Fig. 4. Root length of wheat (*Triticum aestivum*) exposed in hydroponics to different types of CNTs (DWCNTs non-functionalized (DWCNTs nf), DWCNTs functionalized (DWCNTs f) and MWCNTs (Larue et al. unpublished data). Stars indicate significant difference ($p < 0.05$). (A colour version of this figure can be viewed online.)

to understand the controversial results.

Conclusion on toxicity and behaviour of CNTs is difficult to reach due to the different points highlighted earlier in the text. In toxicology, more studies are available and authors hypothesised that short CNTs (30–100 nm) circulate more easily due to their size and are less toxic because they are eliminated easily. Longer CNTs were compared to asbestos. This category may be defined as the most toxic because CNTs can enter into organisms but they cannot be eliminated so they may have toxic effects [9,117]. However, in ecotoxicology, this conclusion is not so obvious.

As the CNT production and uses are expected to be still increasing, their spreading into the environment will keep expanding. It is thus essential to better evaluate CNT behaviour and impacts on ecosystems. More studies are urgently needed to understand mechanistic pathways of penetration and biodistribution of CNTs in plants, microorganisms and macroorganisms in order to allow, if possible, a safe use of CNTs. It is also essential to assess the influence of physico-chemical parameters of CNTs on their impacts. Knowing the effects of these parameters will allow creating CNTs "safer by design".

References

[1] European Commission, Commission Européenne - COMMUNIQUES DE PRESSE - Communiqué de presse - Qu'est-ce qu'un «nanomatériau»?, 2016. http://europa.eu/rapid/press-release_IP-11-1202_fr.htm. (Accessed 4 January 2017).

[2] M.E. Vance, T. Kuiken, E.P. Vejerano, S.P. McGinnis, M.F. Hochella Jr., D. Rejeski, M.S. Hull, Nanotechnology in the real world: redeveloping the nanomaterial consumer products inventory, Beilstein J. Nanotechnol. 6 (2015) 1769–1780, http://dx.doi.org/10.3762/bjnano.6.181.

[3] S. Iijima, Helical microtubules of graphitic carbon, Nature 354 (1991) 56–58, http://dx.doi.org/10.1038/354056a0.

[4] M.S. Dresselhaus, G. Dresselhaus, P. Avouris, Carbon Nanotubes: Synthesis, Structure, Properties, and Applications, Springer Science & Business Media, 2003.

[5] M. Terrones, Carbon nanotubes: synthesis and properties, electronic devices and other emerging applications, Int. Mater. Rev. 49 (2004) 325–377, http://dx.doi.org/10.1179/174328004X5655.

[6] ANSES, R-Nano.fr, 2016. https://www.r-nano.fr/. (Accessed 4 January 2017).

[7] F. Gottschalk, T. Sun, B. Nowack, Environmental concentrations of engineered nanomaterials: review of modeling and analytical studies, Environ.

Pollut. Barking Essex 1987 (181) (2013) 287–300, http://dx.doi.org/10.1016/j.envpol.2013.06.003.
[8] C. Bertrand, Nanomatériaux à travers un gradient de salinité: exposition et effets écotoxicologiques au cours de leur cycle de vie, 2016.
[9] K. Donaldson, C.A. Poland, F.A. Murphy, M. MacFarlane, T. Chernova, A. Schinwald, Pulmonary toxicity of carbon nanotubes and asbestos — similarities and differences, Adv. Drug Deliv. Rev. 65 (2013) 2078–2086, http://dx.doi.org/10.1016/j.addr.2013.07.014.
[10] T. Kasai, Y. Umeda, M. Ohnishi, H. Kondo, T. Takeuchi, S. Aiso, T. Nishizawa, M. Matsumoto, S. Fukushima, Thirteen-week study of toxicity of fiber-like multi-walled carbon nanotubes with whole-body inhalation exposure in rats, Nanotoxicology 9 (2015) 413–422, http://dx.doi.org/10.3109/17435390.2014.933903.
[11] A. Kahru, H.-C. Dubourguier, From ecotoxicology to nanoecotoxicology, Toxicology 269 (2010) 105–119, http://dx.doi.org/10.1016/j.tox.2009.08.016.
[12] A.D. Maynard, R.J. Aitken, T. Butz, V. Colvin, K. Donaldson, G. Oberdörster, M.A. Philbert, J. Ryan, A. Seaton, V. Stone, S.S. Tinkle, L. Tran, N.J. Walker, D.B. Warheit, Safe handling of nanotechnology, Nature 444 (2006) 267–269, http://dx.doi.org/10.1038/444267a.
[13] M.F. Yu, O. Lourie, M.J. Dyer, K. Moloni, T.F. Kelly, R.S. Ruoff, Strength and breaking mechanism of multiwalled carbon nanotubes under tensile load, Science 287 (2000) 637–640.
[14] C.-M. Chang, Y.-L. Liu, Functionalization of multi-walled carbon nanotubes with non-reactive polymers through an ozone-mediated process for the preparation of a wide range of high performance polymer/carbon nanotube composites, Carbon 48 (2010) 1289–1297, http://dx.doi.org/10.1016/j.carbon.2009.12.002.
[15] Z. Yao, C.L. Kane, C. Dekker, High-field electrical transport in single-wall carbon nanotubes, Phys. Rev. Lett. 84 (2000) 2941–2944, http://dx.doi.org/10.1103/PhysRevLett.84.2941.
[16] S. Berber, Y.-K. Kwon, D. Tománek, Unusually high thermal conductivity of carbon nanotubes, Phys. Rev. Lett. 84 (2000) 4613–4616, http://dx.doi.org/10.1103/PhysRevLett.84.4613.
[17] C. Laurent, E. Flahaut, A. Peigney, A. Rousset, Metal nanoparticles for the catalytic synthesis of carbon nanotubes, New J. Chem. 22 (1998) 1229–1237, http://dx.doi.org/10.1039/a801991f.
[18] M. Monthioux, P. Serp, E. Flahaut, M. Razafinimanana, C. Laurent, A. Peigney, W. Bacsa, J.-M. Broto, Introduction to carbon nanotubes, in: B.B. Prof (Ed.), Springer Handb. Nanotechnol, Springer Berlin Heidelberg, 2007, pp. 43–112. http://link.springer.com/referenceworkentry/10.1007/978-3-540-29857-1_3. (Accessed 25 February 2017).
[19] P. Landois, Synthèse, fonctionnalisation et impact sur l'environnement de nanotubes de carbone, Toulouse 3, 2008. http://www.theses.fr/2008TOU30175. (Accessed 6 April 2017).
[20] C. Larue, M. Pinault, B. Czarny, D. Georgin, D. Jaillard, N. Bendiab, M. Mayne-L'Hermite, F. Taran, V. Dive, M. Carrière, Quantitative evaluation of multi-walled carbon nanotube uptake in wheat and rapeseed, J. Hazard. Mater. 227–228 (2012) 155–163, http://dx.doi.org/10.1016/j.jhazmat.2012.05.033.
[21] T.A. Ratnikova, R. Podila, A.M. Rao, A.G. Taylor, Tomato seed coat permeability to selected carbon nanomaterials and enhancement of germination and seedling growth, Sci. World J. 2015 (2015), e419215, http://dx.doi.org/10.1155/2015/419215.
[22] W. Wenseleers, I.I. Vlasov, E. Goovaerts, E.D. Obraztsova, A.S. Lobach, A. Bouwen, Efficient isolation and solubilization of pristine single-walled nanotubes in bile salt micelles, Adv. Funct. Mater 14 (2004) 1105–1112, http://dx.doi.org/10.1002/adfm.200400130.
[23] V. Datsyuk, P. Landois, J. Fitremann, A. Peigney, A.M. Galibert, B. Soula, E. Flahaut, Double-walled carbon nanotube dispersion via surfactant substitution, J. Mater. Chem. 19 (2009) 2729–2736.
[24] J.-M. Bonard, M. Croci, C. Klinke, R. Kurt, O. Noury, N. Weiss, Carbon nanotube films as electron field emitters, Carbon 40 (2002) 1715–1728, http://dx.doi.org/10.1016/S0008-6223(02)00011-8.
[25] Z. Liu, G. Yang, Y.Z. Lee, D. Bordelon, J. Lu, O. Zhou, Carbon nanotube based microfocus field emission x-ray source for microcomputed tomography, Appl. Phys. Lett. 89 (2006), http://dx.doi.org/10.1063/1.2345829.
[26] H. Dai, J.H. Hafner, A.G. Rinzler, D.T. Colbert, R.E. Smalley, Nanotubes as nanoprobes in scanning probe microscopy, Nature 384 (1996) 147–150, http://dx.doi.org/10.1038/384147a0.
[27] C.V. Nguyen, Q. Ye, M. Meyyappan, Carbon nanotube tips for scanning probe microscopy: fabrication and high aspect ratio nanometrology, Meas. Sci. Technol. 16 (2005) 2138, http://dx.doi.org/10.1088/0957-0233/16/11/003.
[28] C.L. Cheung, J.H. Hafner, C.M. Lieber, Carbon nanotube atomic force microscopy tips: direct growth by chemical vapor deposition and application to high-resolution imaging, Proc. Natl. Acad. Sci. 97 (2000) 3809–3813.
[29] Z. Xu, F. Fang, S. Dong, Carbon Nanotube AFM Probe Technology, 2011, http://dx.doi.org/10.5772/17350.
[30] P. Zhai, J.A. Isaacs, M.J. Eckelman, Net energy benefits of carbon nanotube applications, Appl. Energy 173 (2016) 624–634, http://dx.doi.org/10.1016/j.apenergy.2016.04.001.
[31] A. Bianco, K. Kostarelos, C.D. Partidos, M. Prato, Biomedical applications of functionalised carbon nanotubes, Chem. Commun. Camb. Engl. (2005) 571–577, http://dx.doi.org/10.1039/b410943k.
[32] S. Fiorito, E. Flahaut, S. Rapino, F. Paolucci, F. Andreola, N. Moroni, E. Pittaluga, M. Zonfrillo, G. Valenti, A. Mastrofrancesco, F. Groppi, E. Sabbioni, E. Bakalis, F. Zerbetto, A. Serafino, Redox active Double Wall Carbon Nanotubes show intrinsic anti-proliferative effects and modulate autophagy in cancer cells, Carbon 78 (2014) 589–600, http://dx.doi.org/10.1016/j.carbon.2014.07.046.
[33] L. García-Hevia, R. Valiente, J.L. Fernández-Luna, E. Flahaut, L. Rodríguez-Fernández, J.C. Villegas, J. González, M.L. Fanarraga, Inhibition of cancer cell migration by multiwalled carbon nanotubes, Adv. Healthc. Mater 4 (2015) 1640–1644, http://dx.doi.org/10.1002/adhm.201500252.
[34] M.F. Serag, N. Kaji, M. Tokeshi, A. Bianco, Y. Baba, The plant cell uses carbon nanotubes to build tracheary elements, Integr. Biol. Quant. Biosci. Nano Macro 4 (2012) 127–131, http://dx.doi.org/10.1039/c2ib00135g.
[35] A. Gogos, K. Knauer, T.D. Bucheli, Nanomaterials in plant protection and fertilization: current state, foreseen applications, and research priorities, J. Agric. Food Chem. 60 (2012) 9781–9792, http://dx.doi.org/10.1021/jf302154y.
[36] V.K.K. Upadhyayula, S. Deng, M.C. Mitchell, G.B. Smith, Application of carbon nanotube technology for removal of contaminants in drinking water: a review, Sci. Total Environ. 408 (2009) 1–13, http://dx.doi.org/10.1016/j.scitotenv.2009.09.027.
[37] X. Gui, H. Li, K. Wang, J. Wei, Y. Jia, Z. Li, L. Fan, A. Cao, H. Zhu, D. Wu, Recyclable carbon nanotube sponges for oil absorption, Acta Mater 59 (2011) 4798–4804, http://dx.doi.org/10.1016/j.actamat.2011.04.022.
[38] B. Nowack, R.M. David, H. Fissan, H. Morris, J.A. Shatkin, M. Stintz, R. Zepp, D. Brouwer, Potential release scenarios for carbon nanotubes used in composites, Environ. Int. 59 (2013) 1–11, http://dx.doi.org/10.1016/j.envint.2013.04.003.
[39] J. Kolosnjaj-Tabi, J. Just, K.B. Hartman, Y. Laoudi, S. Boudjemaa, D. Alloyeau, H. Szwarc, L.J. Wilson, F. Moussa, Anthropogenic carbon nanotubes found in the airways of parisian children, EBioMedicine 2 (2015) 1697–1704, http://dx.doi.org/10.1016/j.ebiom.2015.10.012.
[40] A. Caballero-Guzman, T. Sun, B. Nowack, Flows of engineered nanomaterials through the recycling process in Switzerland, Waste Manag. 36 (2015) 33–43, http://dx.doi.org/10.1016/j.wasman.2014.11.006.
[41] F. Gottschalk, C. Lassen, J. Kjoelholt, F. Christensen, B. Nowack, Modeling flows and concentrations of nine engineered nanomaterials in the Danish environment, Int. J. Environ. Res. Public. Health 12 (2015) 5581–5602, http://dx.doi.org/10.3390/ijerph120505581.
[42] V.K.K. Upadhyayula, D.E. Meyer, M.A. Curran, M.A. Gonzalez, Life cycle assessment as a tool to enhance the environmental performance of carbon nanotube products: a review, J. Clean. Prod. 26 (2012) 37–47, http://dx.doi.org/10.1016/j.jclepro.2011.12.018.
[43] L. Schlagenhauf, B.T.T. Chu, J. Buha, F. Nüesch, J. Wang, Release of carbon nanotubes from an epoxy-based nanocomposite during an abrasion process, Environ. Sci. Technol. 46 (2012) 7366–7372, http://dx.doi.org/10.1021/es300320y.
[44] T.Y. Sun, N.A. Bornhöft, K. Hungerbühler, B. Nowack, Dynamic probabilistic modeling of environmental emissions of engineered nanomaterials, Environ. Sci. Technol. 50 (2016) 4701–4711, http://dx.doi.org/10.1021/acs.est.5b05828.
[45] B. Czarny, D. Georgin, F. Berthon, G. Plastow, M. Pinault, G. Patriarche, A. Thuleau, M.M. L'Hermite, F. Taran, V. Dive, Carbon nanotube translocation to distant organs after pulmonary exposure: insights from in situ 14C-radiolabeling and tissue radioimaging, ACS Nano 8 (2014) 5715–5724, http://dx.doi.org/10.1021/nn500475u.
[46] A. Mottier, F. Mouchet, C. Laplanche, S. Cadarsi, L. Lagier, J.-C. Arnault, H.A. Girard, V. León, E. Vázquez, C. Sarrieu, É. Pinelli, L. Gauthier, E. Flahaut, Surface area of carbon nanoparticles: a dose metric for a more realistic ecotoxicological assessment, Nano Lett. 16 (2016) 3514–3518, http://dx.doi.org/10.1021/acs.nanolett.6b00348.
[47] G. Fagerlund, Determination of specific surface by the BET method, Matér. Constr. 6 (1973) 239–245, http://dx.doi.org/10.1007/BF02479039.
[48] F. Bourdiol, F. Mouchet, A. Perrault, I. Fourquaux, L. Datas, C. Gancet, J.-C. Boutonnet, E. Pinelli, L. Gauthier, E. Flahaut, Biocompatible polymer-assisted dispersion of multi walled carbon nanotubes in water, application to the investigation of their ecotoxicity using Xenopus laevis amphibian larvae, Carbon 54 (2013) 175–191, http://dx.doi.org/10.1016/j.carbon.2012.11.024.
[49] C. Herrero-Latorre, J. Álvarez-Méndez, J. Barciela-García, S. García-Martín, R.M. Peña-Crecente, Characterization of carbon nanotubes and analytical methods for their determination in environmental and biological samples: a review, Anal. Chim. Acta 853 (2015) 77–94, http://dx.doi.org/10.1016/j.aca.2014.10.008.
[50] J. Barbillat, D. Bougeard, G. Buntinx, M. Delhaye, P. Dhamelincourt, F. Fillaux, Spectrométrie raman, Tech. Ing. 2 (1999) 2865.
[51] C. Lamprecht, I. Liashkovich, V. Neves, J. Danzberger, E. Heister, M. Rangl, H.M. Coley, J. McFadden, E. Flahaut, H.J. Gruber, P. Hinterdorfer, F. Kienberger, A. Ebner, AFM imaging of functionalized carbon nanotubes on biological membranes, Nanotechnology 20 (2009), 434001, http://dx.doi.org/10.1088/0957-4484/20/43/434001.
[52] M. Alimohammadi, Y. Xu, D. Wang, A.S. Biris, M.V. Khodakovskaya, Physiological responses induced in tomato plants by a two-component nanostructural system composed of carbon nanotubes conjugated with quantum dots and its in vivo multimodal detection, Nanotechnology 22 (2011), 295101, http://dx.doi.org/10.1088/0957-4484/22/29/295101.
[53] A. Mondal, R. Basu, S. Das, P. Nandy, Beneficial role of carbon nanotubes on

mustard plant growth: an agricultural prospect, J. Nanoparticle Res. 13 (2011) 4519, http://dx.doi.org/10.1007/s11051-011-0406-z.
[54] P.V. Jena, Y. Shamay, J. Shah, D. Roxbury, N. Paknejad, D.A. Heller, Photoluminescent carbon nanotubes interrogate the permeability of multicellular tumor spheroids, Carbon 97 (2016) 99–109, http://dx.doi.org/10.1016/j.carbon.2015.08.024.
[55] P. Lutsyk, R. Arif, J. Hruby, A. Bukivskyi, O. Vinijchuk, M. Shandura, V. Yakubovskyi, Y. Kovtun, G.A. Rance, M. Fay, Y. Piryatinski, O. Kachkovsky, A. Verbitsky, A. Rozhin, A sensing mechanism for the detection of carbon nanotubes using selective photoluminescent probes based on ionic complexes with organic dyes, Light Sci. Appl. 5 (2016), e16028, http://dx.doi.org/10.1038/lsa.2016.28.
[56] K. Doudrick, P. Herckes, P. Westerhoff, Detection of carbon nanotubes in environmental matrices using programmed thermal analysis, Environ. Sci. Technol. 46 (2012) 12246–12253, http://dx.doi.org/10.1021/es300804f.
[57] F. Irin, B. Shrestha, J.E. Cañas, M.A. Saed, M.J. Green, Detection of carbon nanotubes in biological samples through microwave-induced heating, Carbon 50 (2012) 4441–4449, http://dx.doi.org/10.1016/j.carbon.2012.05.022.
[58] F. Bourdiol, D. Dubuc, K. Grenier, F. Mouchet, L. Gauthier, E. Flahaut, Quantitative detection of carbon nanotubes in biological samples by an original method based on microwave permittivity measurements, Carbon 81 (2015) 535–545, http://dx.doi.org/10.1016/j.carbon.2014.09.086.
[59] S. Chen, C. Xiong, H. Liu, Q. Wan, J. Hou, Q. He, A. Badu-Tawiah, Z. Nie, Mass spectrometry imaging reveals the sub-organ distribution of carbon nanomaterials, Nat. Nanotechnol. 10 (2015) 176–182, http://dx.doi.org/10.1038/nnano.2014.282.
[60] B.R. Smith, E.E.B. Ghosn, H. Rallapalli, J.A. Prescher, T. Larson, L.A. Herzenberg, S.S. Gambhir, Selective uptake of single-walled carbon nanotubes by circulating monocytes for enhanced tumour delivery, Nat. Nanotechnol. 9 (2014) 481–487, http://dx.doi.org/10.1038/nnano.2014.62.
[61] M.V. Khodakovskaya, K. de Silva, D.A. Nedosekin, E. Dervishi, A.S. Biris, E.V. Shashkov, E.I. Galanzha, V.P. Zharov, Complex genetic, photothermal, and photoacoustic analysis of nanoparticle-plant interactions, Proc. Natl. Acad. Sci. U. S. A. 108 (2011) 1028–1033, http://dx.doi.org/10.1073/pnas.1008856108.
[62] P. Jackson, N.R. Jacobsen, A. Baun, R. Birkedal, D. Kühnel, K.A. Jensen, U. Vogel, H. Wallin, Bioaccumulation and ecotoxicity of carbon nanotubes, Chem. Cent. J. 7 (2013) 154, http://dx.doi.org/10.1186/1752-153X-7-154.
[63] D.P. Jaisi, M. Elimelech, Single-Walled carbon nanotubes exhibit limited transport in soil columns, Environ. Sci. Technol. 43 (2009) 9161–9166, http://dx.doi.org/10.1021/es901927y.
[64] D. Kasel, S.A. Bradford, J. Šimůnek, T. Pütz, H. Vereecken, E. Klumpp, Limited transport of functionalized multi-walled carbon nanotubes in two natural soils, Environ. Pollut. 180 (2013) 152–158, http://dx.doi.org/10.1016/j.envpol.2013.05.031.
[65] Y. Lu, K. Yang, D. Lin, Transport of surfactant-facilitated multiwalled carbon nanotube suspensions in columns packed with sized soil particles, Environ. Pollut. 192 (2014) 36–43, http://dx.doi.org/10.1016/j.envpol.2014.05.008.
[66] G. Cornelis, K. Hund-Rinke, T. Kuhlbusch, N. van den Brink, C. Nickel, Fate and bioavailability of engineered nanoparticles in soils: a review, Crit. Rev. Environ. Sci. Technol. 44 (2014) 2720–2764, http://dx.doi.org/10.1080/10643389.2013.829767.
[67] T. Hiroi, S. Ata, M. Shibayama, Transitions of aggregation states for concentrated carbon nanotube dispersion, J. Phys. Chem. C 120 (2016) 5776–5782, http://dx.doi.org/10.1021/acs.jpcc.5b12464.
[68] D. Bouchard, X. Chang, I. Chowdhury, Heteroaggregation of multiwalled carbon nanotubes with sediments, Environ. Nanotechnol. Monit. Manag. 4 (2015) 42–50, http://dx.doi.org/10.1016/j.enmm.2015.07.001.
[69] B. Shrestha, T.A. Anderson, V. Acosta-Martinez, P. Payton, J.E. Cañas-Carrell, The influence of multiwalled carbon nanotubes on polycyclic aromatic hydrocarbon (PAH) bioavailability and toxicity to soil microbial communities in alfalfa rhizosphere, Ecotoxicol. Environ. Saf. 116 (2015) 143–149, http://dx.doi.org/10.1016/j.ecoenv.2015.03.005.
[70] R. De La Torre-Roche, J. Hawthorne, Y. Deng, B. Xing, W. Cai, L.A. Newman, Q. Wang, X. Ma, H. Hamdi, J.C. White, Multiwalled carbon nanotubes and C60 fullerenes differentially impact the accumulation of weathered pesticides in four agricultural plants, Environ. Sci. Technol. 47 (2013) 12539–12547, http://dx.doi.org/10.1021/es4034809.
[71] H. Chung, Y. Son, T.K. Yoon, S. Kim, W. Kim, The effect of multi-walled carbon nanotubes on soil microbial activity, Ecotoxicol. Environ. Saf. 74 (2011) 569–575, http://dx.doi.org/10.1016/j.ecoenv.2011.01.004.
[72] L. Jin, Y. Son, T.K. Yoon, Y.J. Kang, W. Kim, H. Chung, High concentrations of single-walled carbon nanotubes lower soil enzyme activity and microbial biomass, Ecotoxicol. Environ. Saf. 88 (2013) 9–15, http://dx.doi.org/10.1016/j.ecoenv.2012.10.031.
[73] L. Jin, Y. Son, J.L. DeForest, Y.J. Kang, W. Kim, H. Chung, Single-walled carbon nanotubes alter soil microbial community composition, Sci. Total Environ. 466–467 (2014) 533–538, http://dx.doi.org/10.1016/j.scitotenv.2013.07.035.
[74] Y. Ge, J.H. Priester, M. Mortimer, C.H. Chang, Z. Ji, J.P. Schimel, P.A. Holden, Long-term effects of multiwalled carbon nanotubes and graphene on microbial communities in dry soil, Environ. Sci. Technol. 50 (2016) 3965–3974, http://dx.doi.org/10.1021/acs.est.5b05620.
[75] D.F. Rodrigues, D.P. Jaisi, M. Elimelech, Toxicity of functionalized single-walled carbon nanotubes on soil microbial communities: implications for nutrient cycling in soil, Environ. Sci. Technol. 47 (2013) 625–633, http://dx.doi.org/10.1021/es304002q.
[76] J. Shan, R. Ji, Y. Yu, Z. Xie, X. Yan, Biochar, activated carbon, and carbon nanotubes have different effects on fate of 14C-catechol and microbial community in soil, Sci. Rep. 5 (2015), 16000., http://dx.doi.org/10.1038/srep16000.
[77] E.J. Petersen, Q. Huang, W.J. Weber, Bioaccumulation of radio-labeled carbon nanotubes by Eisenia foetida, Environ. Sci. Technol. 42 (2008) 3090–3095.
[78] E.J. Petersen, R.A. Pinto, P.F. Landrum, J. Weber Walter, Influence of carbon nanotubes on pyrene bioaccumulation from contaminated soils by earthworms, Environ. Sci. Technol. 43 (2009) 4181–4187, http://dx.doi.org/10.1021/es803023a.
[79] E.J. Petersen, R.A. Pinto, L. Zhang, Q. Huang, P.F. Landrum, W.J. Weber, Effects of polyethyleneimine-mediated functionalization of multi-walled carbon nanotubes on earthworm bioaccumulation and sorption by soils, Environ. Sci. Technol. 45 (2011) 3718–3724, http://dx.doi.org/10.1021/es103004r.
[80] J.J. Scott-Fordsmand, P.H. Krogh, M. Schaefer, A. Johansen, The toxicity testing of double-walled nanotubes-contaminated food to Eisenia veneta earthworms, Ecotoxicol. Environ. Saf. 71 (2008) 616–619, http://dx.doi.org/10.1016/j.ecoenv.2008.04.011.
[81] C. Hu, Y. Cai, W. Wang, Y. Cui, M. Li, Toxicological effects of multi-walled carbon nanotubes adsorbed with nonylphenol on earthworm Eisenia fetida, Environ. Sci. Process. Impacts 15 (2013) 2125–2130, http://dx.doi.org/10.1039/c3em00376k.
[82] A. Calisi, A. Grimaldi, A. Leomanni, M.G. Lionetto, F. Dondero, T. Schettino, Multibiomarker response in the earthworm Eisenia fetida as tool for assessing multi-walled carbon nanotube ecotoxicity, Ecotoxicol. Lond. Engl. 25 (2016) 677–687, http://dx.doi.org/10.1007/s10646-016-1626-x.
[83] S. Li, F. Irin, F.O. Atore, M.J. Green, J.E. Cañas-Carrell, Determination of multi-walled carbon nanotube bioaccumulation in earthworms measured by a microwave-based detection technique, Sci, Total Environ. 445–446 (2013) 9–13, http://dx.doi.org/10.1016/j.scitotenv.2012.12.037.
[84] H.-J. Eom, C.P. Roca, J.-Y. Roh, N. Chatterjee, J.-S. Jeong, I. Shim, H.-M. Kim, P.-J. Kim, K. Choi, F. Giralt, J. Choi, A systems toxicology approach on the mechanism of uptake and toxicity of MWCNT in Caenorhabditis elegans, Chem. Biol. Interact. 239 (2015) 153–163, http://dx.doi.org/10.1016/j.cbi.2015.06.031.
[85] L. Zhang, C. Hu, W. Wang, F. Ji, Y. Cui, M. Li, Acute toxicity of multi-walled carbon nanotubes, sodium pentachlorophenate, and their complex on earthworm Eisenia fetida, Ecotoxicol. Environ. Saf. 103 (2014) 29–35, http://dx.doi.org/10.1016/j.ecoenv.2014.01.041.
[86] M.C. Martínez-Ballesta, L. Zapata, N. Chalbi, M. Carvajal, Multiwalled carbon nanotubes enter broccoli cells enhancing growth and water uptake of plants exposed to salinity, J. Nanobiotechnol. 14 (2016) 42, http://dx.doi.org/10.1186/s12951-016-0199-4.
[87] Y. Hao, F. Yu, R. Lv, C. Ma, Z. Zhang, Y. Rui, L. Liu, W. Cao, B. Xing, Carbon nanotubes filled with different ferromagnetic alloys affect the growth and development of rice seedlings by changing the C: N ratio and plant hormones concentrations, PLoS One 11 (2016), e0157264, http://dx.doi.org/10.1371/journal.pone.0157264.
[88] M. Khodakovskaya, E. Dervishi, M. Mahmood, Y. Xu, Z. Li, F. Watanabe, A.S. Biris, Carbon nanotubes are able to penetrate plant seed coat and dramatically affect seed germination and plant growth, ACS Nano 3 (2009) 3221–3227, http://dx.doi.org/10.1021/nn900887m.
[89] M.H. Lahiani, E. Dervishi, I. Ivanov, J. Chen, M. Khodakovskaya, Comparative study of plant responses to carbon-based nanomaterials with different morphologies, Nanotechnology 27 (2016) 265102, http://dx.doi.org/10.1088/0957-4484/27/26/265102.
[90] M.H. Lahiani, E. Dervishi, J. Chen, Z. Nima, A. Gaume, A.S. Biris, M.V. Khodakovskaya, Impact of carbon nanotube exposure to seeds of valuable crops, ACS Appl. Mater. Interfaces 5 (2013) 7965–7973, http://dx.doi.org/10.1021/am402052x.
[91] D.K. Tiwari, N. Dasgupta-Schubert, L.M.V. Cendejas, J. Villegas, L.C. Montoya, S.E.B. García, Interfacing carbon nanotubes (CNT) with plants: enhancement of growth, water and ionic nutrient uptake in maize (Zea mays) and implications for nanoagriculture, Appl. Nanosci. 4 (2013) 577–591, http://dx.doi.org/10.1007/s13204-013-0236-7.
[92] G. Chen, J. Qiu, Y. Liu, R. Jiang, S. Cai, Y. Liu, F. Zhu, F. Zeng, T. Luan, G. Ouyang, Carbon nanotubes act as contaminant carriers and translocate within plants, Sci. Rep. 5 (2015), http://dx.doi.org/10.1038/srep15682.
[93] C. Lin, B. Fugetsu, Y. Su, F. Watari, Studies on toxicity of multi-walled carbon nanotubes on Arabidopsis T87 suspension cells, J. Hazard. Mater 170 (2009) 578–583, http://dx.doi.org/10.1016/j.jhazmat.2009.05.025.
[94] P. Miralles, E. Johnson, T.L. Church, A.T. Harris, Multiwalled carbon nanotubes in alfalfa and wheat: toxicology and uptake, J. R. Soc. Interface R. Soc. 9 (2012) 3514–3527, http://dx.doi.org/10.1098/rsif.2012.0535.
[95] P. Begum, R. Ikhtiari, B. Fugetsu, M. Matsuoka, T. Akasaka, F. Watari, Phytotoxicity of multi-walled carbon nanotubes assessed by selected plant species in the seedling stage, Appl. Surf. Sci. 262 (2012) 120–124, http://dx.doi.org/10.1016/j.apsusc.2012.03.028.
[96] E. Smirnova, A. Gusev, O. Zaytseva, O. Sheina, A. Tkachev, E. Kuznetsova, E. Lazareva, G. Onishchenko, A. Feofanov, M. Kirpichnikov, Uptake and accumulation of multiwalled carbon nanotubes change the morphometric and biochemical characteristics of Onobrychis arenaria seedlings, Front. Chem. Sci. Eng. 6 (2012) 132–138, http://dx.doi.org/10.1007/s11705-012-

1290-5.
[97] G. Zhai, S.M. Gutowski, K.S. Walters, B. Yan, J.L. Schnoor, Charge, size, and cellular selectivity for multiwall carbon nanotubes by maize and soybean, Environ. Sci. Technol. 49 (2015) 7380−7390, http://dx.doi.org/10.1021/acs.est.5b01145.
[98] A.M. Cano, K. Kohl, S. Deleon, P. Payton, F. Irin, M. Saed, S.A. Shah, M.J. Green, J.E. Cañas-Carrell, Determination of uptake, accumulation, and stress effects in corn (Zea mays L.) grown in single-wall carbon nanotube contaminated soil, Chemosphere 152 (2016) 117−122, http://dx.doi.org/10.1016/j.chemosphere.2016.02.093.
[99] M.F. Serag, N. Kaji, C. Gaillard, Y. Okamoto, K. Terasaka, M. Jabasini, M. Tokeshi, H. Mizukami, A. Bianco, Y. Baba, Trafficking and subcellular localization of multiwalled carbon nanotubes in plant cells, ACS Nano 5 (2011) 493−499, http://dx.doi.org/10.1021/nn102344t.
[100] M.F. Serag, N. Kaji, S. Habuchi, A. Bianco, Y. Baba, Nanobiotechnology meets plant cell biology: carbon nanotubes as organelle targeting nanocarriers, RSC Adv. 3 (2013) 4856−4862, http://dx.doi.org/10.1039/C2RA22766E.
[101] S. Lin, J. Reppert, Q. Hu, J.S. Hudson, M.L. Reid, T.A. Ratnikova, A.M. Rao, H. Luo, P.C. Ke, Uptake, translocation, and transmission of carbon nanomaterials in rice plants, Small Weinh. Bergstr. Ger. 5 (2009) 1128−1132, http://dx.doi.org/10.1002/smll.200801556.
[102] P. Begum, B. Fugetsu, Phytotoxicity of multi-walled carbon nanotubes on red spinach (Amaranthus tricolor L) and the role of ascorbic acid as an antioxidant, J. Hazard. Mater 243 (2012) 212−222, http://dx.doi.org/10.1016/j.jhazmat.2012.10.025.
[103] P. Oleszczuk, I. Jośko, B. Xing, The toxicity to plants of the sewage sludges containing multiwalled carbon nanotubes, J. Hazard. Mater 186 (2011) 436−442, http://dx.doi.org/10.1016/j.jhazmat.2010.11.028.
[104] M.H. Lahiani, J. Chen, F. Irin, A.A. Puretzky, M.J. Green, M.V. Khodakovskaya, Interaction of carbon nanohorns with plants: uptake and biological effects, Carbon 81 (2015) 607−619, http://dx.doi.org/10.1016/j.carbon.2014.09.095.
[105] S. Tripathi, S.K. Sonkar, S. Sarkar, Growth stimulation of gram (Cicer arietinum) plant by water soluble carbon nanotubes, Nanoscale 3 (2011) 1176−1181, http://dx.doi.org/10.1039/c0nr00722f.
[106] H. Villagarcia, E. Dervishi, K. de Silva, A.S. Biris, M.V. Khodakovskaya, Surface chemistry of carbon nanotubes impacts the growth and expression of water channel protein in tomato plants, Small 8 (2012) 2328−2334, http://dx.doi.org/10.1002/smll.201102661.
[107] M.V. Khodakovskaya, B.-S. Kim, J.N. Kim, M. Alimohammadi, E. Dervishi, T. Mustafa, C.E. Cernigla, Carbon nanotubes as plant growth regulators: effects on tomato growth, reproductive system, and soil microbial community, Small Weinh. Bergstr. Ger. 9 (2013) 115−123, http://dx.doi.org/10.1002/smll.201201225.
[108] Y. Zhang, Q. Sun, J. Zhou, S. Masunaga, F. Ma, Reduction in toxicity of wastewater from three wastewater treatment plants to alga (Scenedesmus obliquus) in northeast China, Ecotoxicol. Environ. Saf. 119 (2015) 132−139, http://dx.doi.org/10.1016/j.ecoenv.2015.04.034.
[109] R.A. Taha, M.M. Hassan, E.A. Ibrahim, N.H.A. Baker, E.A. Shaaban, Carbon nanotubes impact on date palm in vitro cultures, Plant Cell Tissue Organ Cult, PCTOC 127 (2016) 525−534, http://dx.doi.org/10.1007/s11240-016-1058-6.
[110] C.-X. Shen, Q.-F. Zhang, J. Li, F.-C. Bi, N. Yao, Induction of programmed cell death in Arabidopsis and rice by single-wall carbon nanotubes, Am. J. Bot. 97 (2010) 1602−1609, http://dx.doi.org/10.3732/ajb.1000073.
[111] H. Hamdi, R. De La Torre-Roche, J. Hawthorne, J.C. White, Impact of non-functionalized and amino-functionalized multiwall carbon nanotubes on pesticide uptake by lettuce (Lactuca sativa L.), Nanotoxicology 9 (2015) 172−180, http://dx.doi.org/10.3109/17435390.2014.907456.
[112] D. Lin, B. Xing, Phytotoxicity of nanoparticles: inhibition of seed germination and root growth, Environ. Pollut. 150 (2007) 243−250, http://dx.doi.org/10.1016/j.envpol.2007.01.016.
[113] J.E. Cañas, M. Long, S. Nations, R. Vadan, L. Dai, M. Luo, R. Ambikapathi, E.H. Lee, D. Olszyk, Effects of functionalized and nonfunctionalized single-walled carbon nanotubes on root elongation of select crop species, Environ. Toxicol. Chem. SETAC 27 (2008) 1922−1931.
[114] M.V. Khodakovskaya, K. de Silva, A.S. Biris, E. Dervishi, H. Villagarcia, Carbon nanotubes induce growth enhancement of tobacco cells, ACS Nano 6 (2012) 2128−2135, http://dx.doi.org/10.1021/nn204643g.
[115] S. Yan, H. Zhang, Y. Huang, J. Tan, P. Wang, Y. Wang, H. Hou, J. Huang, L. Li, Single-wall and multi-wall carbon nanotubes promote rice root growth by eliciting the similar molecular pathways and epigenetic regulation, IET Nanobiotechnol 10 (2016) 222−229, http://dx.doi.org/10.1049/iet-nbt.2015.0046.
[116] S. García-Sánchez, I. Bernales, S. Cristobal, Early response to nanoparticles in the Arabidopsis transcriptome compromises plant defence and root-hair development through salicylic acid signalling, BMC Genomics 16 (2015), http://dx.doi.org/10.1186/s12864-015-1530-4.
[117] M.-C.F. Jaurand, A. Renier, J. Daubriac, Mesothelioma: do asbestos and carbon nanotubes pose the same health risk? Part. Fibre Toxicol. 6 (2009) 16, http://dx.doi.org/10.1186/1743-8977-6-16.
[118] X. Zheng, Y. Su, Y. Chen, R. Wan, M. Li, Y. Wei, H. Huang, Carboxyl-modified single-walled carbon nanotubes negatively affect bacterial growth and denitrification activity, Sci. Rep. 4 (2014) 5653, http://dx.doi.org/10.1038/srep05653.
[119] Z. Tong, M. Bischoff, L.F. Nies, P. Myer, B. Applegate, R.F. Turco, Response of soil microorganisms to as-produced and functionalized single-wall carbon nanotubes (SWNTs), Environ. Sci. Technol. 46 (2012) 13471−13479, http://dx.doi.org/10.1021/es303251r.
[120] A. Simon-Deckers, S. Loo, M. Mayne-L'hermite, N. Herlin-Boime, N. Menguy, C. Reynaud, B. Gouget, M. Carrière, Size-, composition- and shape-dependent toxicological impact of metal oxide nanoparticles and carbon nanotubes toward bacteria, Environ. Sci. Technol. 43 (2009) 8423−8429, http://dx.doi.org/10.1021/es9016975.
[121] Z. Tong, M. Bischoff, L.F. Nies, P. Myer, B. Applegate, R.F. Turco, Response of soil microorganisms to as-produced and functionalized single-wall carbon nanotubes (SWNTs), Environ. Sci. Technol. 46 (2012) 13471−13479, http://dx.doi.org/10.1021/es303251r.
[122] X. Zheng, Y. Su, Y. Chen, R. Wan, M. Li, Y. Wei, H. Huang, Carboxyl-modified single-walled carbon nanotubes negatively affect bacterial growth and denitrification activity, Sci. Rep. 4 (2014) 5653, http://dx.doi.org/10.1038/srep05653.
[123] X. Zheng, Y. Su, Y. Chen, R. Wan, M. Li, Y. Wei, H. Huang, Carboxyl-modified single-walled carbon nanotubes negatively affect bacterial growth and denitrification activity, Sci. Rep. 4 (2014) 5653, http://dx.doi.org/10.1038/srep05653.
[124] Y. Deng, B. Eitzer, J.C. White, B. Xing, Impact of multiwall carbon nanotubes on the accumulation and distribution of carbamazepine in collard greens (Brassica oleracea), Environ. Sci. Nano (2017), http://dx.doi.org/10.1039/C6EN00419A.
[125] J.P. Giraldo, M.P. Landry, S.M. Faltermeier, T.P. McNicholas, N.M. Iverson, A.A. Boghossian, N.F. Reuel, A.J. Hilmer, F. Sen, J.A. Brew, M.S. Strano, Plant nanobionics approach to augment photosynthesis and biochemical sensing, Nat. Mater 13 (2014) 400−408, http://dx.doi.org/10.1038/nmat3890.
[126] M. Hatami, J. Hadian, M. Ghorbanpour, Mechanisms underlying toxicity and stimulatory role of single-walled carbon nanotubes in Hyoscyamus Niger during drought stress simulated by polyethylene glycol, J. Hazard. Mater. 324 (2017) 306−320, http://dx.doi.org/10.1016/j.jhazmat.2016.10.064.
[127] J. Moll, A. Gogos, T.D. Bucheli, F. Widmer, M.G.A. van der Heijden, Effect of nanoparticles on red clover and its symbiotic microorganisms, J. Nanobiotechnol. 14 (2016) 36, http://dx.doi.org/10.1186/s12951-016-0188-7.
[128] S. Park, Y.-J. Ahn, Multi-walled carbon nanotubes and silver nanoparticles differentially affect seed germination, chlorophyll content, and hydrogen peroxide accumulation in carrot (Daucus carota L.), Biocatal. Agric. Biotechnol. 8 (2016) 257−262, http://dx.doi.org/10.1016/j.bcab.2016.09.012.
[129] S. Park, Y.-J. Ahn, Multi-walled carbon nanotubes and silver nanoparticles differentially affect seed germination, chlorophyll content, and hydrogen peroxide accumulation in carrot (Daucus carota L.), Biocatal. Agric. Biotechnol. 8 (2016) 257−262, http://dx.doi.org/10.1016/j.bcab.2016.09.012.
[130] D. Stampoulis, S.K. Sinha, J.C. White, Assay-dependent phytotoxicity of nanoparticles to plants, Environ. Sci. Technol. 43 (2009) 9473−9479, http://dx.doi.org/10.1021/es901695c.
[131] X.-M. Tan, B. Fugetsu, Multi-Walled carbon nanotubes interact with cultured rice cells: evidence of a self-defense response, J. Biomed. Nanotechnol. 3 (2007) 285−288, http://dx.doi.org/10.1166/jbn.2007.035.
[132] X. Tan, C. Lin, B. Fugetsu, Studies of towicity of multi-walled carbon nanotubes on suspension rice cells, Carbon 47 (2009) 3479−3487.
[133] O. Zaytseva, Z. Wang, G. Neumann, Phytotoxicity of carbon nanotubes in soybean as determined by interactions with micronutrients, J. Nanopart Res. 19 (2017) 29, http://dx.doi.org/10.1007/s11051-016-3722-5.
[134] Q. Zhao, C. Ma, J.C. White, O.P. Dhankher, X. Zhang, S. Zhang, B. Xing, Quantitative evaluation of multi-wall carbon nanotube uptake by terrestrial plants, Carbon 114 (2017) 661−670, http://dx.doi.org/10.1016/j.carbon.2016.12.036.
[135] H. Zhang, M. Yue, X. Zheng, C. Xie, H. Zhou, L. Li, Physiological effects of single- and multi-walled carbon nanotubes on rice seedlings, IEEE Trans. Nanobioscience (2017), http://dx.doi.org/10.1109/TNB.2017.2715359.
[136] H. Zhang, Y. Liu, X. Shen, M. Zhang, Y. Yang, S. Tao, X. Wang, Influence of multiwalled carbon nanotubes and sodium dodecyl benzene sulfonate on bioaccumulation and translocation of pyrene and 1-methylpyrene in maize (Zea mays) seedlings, Environ. Pollut. 220 (2017) 1409−1417, http://dx.doi.org/10.1016/j.envpol.2016.10.093.